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Development of a Pharmacokinetic Interaction Model for Co-administration of Simvastatin and Amlodipine

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Summary: A model for drug interaction between amlodipine and simvastatin was developed using concentration data obtained from a multiple-dose study consisting of single- and co-administration of amlodipine and simvastatin conducted in healthy Koreans. Amlodipine concentrations were assumed to influence the clearance of simvastatin and simvastatin acid, which as well as the oral bioavailability was allowed to vary depending on genetic polymorphisms of metabolic enzymes. Covariate effects on drug concentrations were also considered. The developed model yielded a 46% increase in simvastatin bioavailability and a 13% decrease in simvastatin clearance when amlodipine 10 mg was co-administered. When CYP3A4/5 polymorphisms were assessed by a mixture model, extensive metabolizers yielded a decrease in simvastatin bioavailability of 81% and a decrease in simvastatin clearance by 4.6 times as compared to poor metabolizers. Sixty percent of the usual dose was the optimal simvastatin dose that can minimize the interaction with amlodipine 10 mg. Age and weight had significant effects on amlodipine concentrations. In conclusion, this study has quantitatively described the pharmacokinetic interaction between simvastatin and amlodipine using a modeling approach. Given that the two drugs are often prescribed together, the developed model is expected to contribute to more efficient and safer drug treatment when they are co-administered.

Keywords: drug-drug interaction; simvastatin; amlodipine; metabolite; mixture model; population anlyisis

Introduction

Pharmacokinetic drug interactions can occur when at least two drugs are co-administered, typically increasing or decreasing the concentration of the first drug, and these altered concentrations can cause an unexpected adverse drug reaction or therapeutic failure.[1)] A notable metabolic enzyme system involved in drug interactions is the cytochrome P450 oxidase family (CYP450), which, when inhibited or induced by drug interactions, affects the plasma concentrations of a drug. Competitive inhibition is a form of enzyme inhibition whereby binding of an inhibitor to the active site on the enzyme prevents binding of the substrate and vice versa.[2)] When two or more drugs that use a common enzymatic system are administered concomitantly, they can interact with each other in a competitive manner.

Simvastatin is a hydroxyl-methylglutaryl coenzyme A (HMG-CoA) reductase inhibitor used to control hypercholesterolemia.[3)] Simvastatin lowers cholesterol levels in the blood by blocking an enzyme system that creates cholesterol. Simvastatin is metabolized through a complex system including acid/lactone interconversion *via* various pathways. Simvastatin, which is an inactive lactone, is converted to its active form, simvastatin acid, by esterases, paraoxonases and non-enzymatic hydrolysis. Simvastatin acid is converted back to simvastatin *via* the acyl glucuronide intermediate and the CoASH-dependent pathway.[3)] Metabolic elimination of both simvastatin and simvastatin acid occur *via* cytochrome P450 3A4/5 (CYP3A4/5). It has been reported that the CYP3A5 genetic polymorphism influences the pharmacokinetics of simvastatin.[4)]

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Amlodipine is a long-acting calcium channel blocker [dihydropyridine (DHP) class] used as an anti-hypertensive and in the treatment of angina.[5,6] In hypertension, like other calcium channel blockers, amlodipine reduces blood pressure by relaxing the smooth muscle in the arterial wall and decreasing total peripheral resistance and in angina it increases blood flow to the heart muscle.

In reality, simvastatin is often co-administered with amlodipine for the treatment of hyperlipidemia accompanied by hypertension. However, CYP3A is a known major enzyme subfamily responsible for the metabolism of amlodipine as well as simvastatin and simvastatin acid.[3,7] It has been reported that the concentration of simvastatin increases when co-administered with amlodipine, which can competitively inhibit the metabolic activity of CYP3A4/5. On the other hand, it is known that amlodipine concentration is not influenced by simvastatin when the two drugs are co-administered.[8]

Thus, when simvastatin and amlodipine are co-administered, amlodipine can compete with simvastatin in the same metabolic pathway, altering simvastatin metabolism and consequently raising the risk of an adverse drug reaction due to increased blood concentrations of simvastatin. To date, however, population pharmacokinetic analyses of drug-drug interactions between simvastatin and amlodipine have not been conducted.

The objectives of this study were (i) to develop a population model to describe the pharmacokinetics of simvastatin, simvastatin acid, amlodipine, and their drug interactions from data obtained from a pharmacokinetic interaction study between simvastatin and amlodipine; and (ii) to propose the optimal dose of simvastatin needed to reduce the risk of an adverse drug reaction when the two drugs are given together.

Materials and Methods

Pharamcokinetic data: The pharmacokinetic data were obtained from a randomized, open-label, cross-over study conducted in healthy Korean male volunteers in 2009 to investigate the pharmacokinetics and drug interactions between simvastatin and amlodipine.

The subjects were given a Zocor tablet 40 mg (MSD Co., Ltd.; simvastatin 40 mg) only (single-administration) or a Zocor tablet 40 mg and a Norvasc tablet 10 mg (Pfizer Co., Ltd.; amlodipine 10 mg) concomitantly (co-administration), each formulation given *q.d.* for nine consecutive days, with a 14-day washout period. Blood samples were collected on Day 9 before and 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 8, 10, 12, 16, 24, 48, 72, 96, 144, and 216 h after dosing for co-administration or only up to 24 h for single-administration.

The study was approved by the Institutional Review Board of Yonsei University Severance Hospital (Seoul, Korea) and performed in accordance with the Declaration of Helsinki.

Sample assay: Plasma concentrations were quantified by LC-MS/MS using the Agilent 1200 system (Agilent, Santa Clara, CA) for HPLC and the 4000QTAP/API3200 system (Applied Biosystems/MDS SCIEX, Foster City, CA) for MS/MS. Detection was achieved using MRM mode of positive electrospray ionization (ESI^+).

Each concentration of simvastatin, simvastatin acid and amlodipine was determined by spiking 0.5 mL of plasma with lovastatin 15 µL, lovastatin acid 15 µL, and nimodipine 50 µL respectively as an internal standard, and was extracted by protein precipitation. They were detected by mass spectrometry,[9–12] operating under the following conditions of mass transitions: 419→285 m/z for simvastatin, 437→303 m/z for simvastatin acid, 405→199 m/z for lovastatin, 423→285 m/z for lovastatin acid, 409→238 m/z for amlodipine, and 419→343 m/z for nimodipine. The lower limit of quantification (LLOQ) was 0.1 ng/mL for simvastatin and simvastatin acid and 0.5 ng/mL for amlodipine, with assay signal-to-noise (S/N) ratio >10.

The calibration curves were linear over the range of 0.2–50 ng/mL, 0.1–25 ng/mL, and 0.2–20 ng/mL for simvastatin, simvastatin acid, and amlodipine, respectively. They were developed using the following criteria: (1) the mean value should be within ±15% of the theoretical value (2) the precision should not exceed 15% in coefficient of variation and (3) the correlation coefficient should be greater than 0.95.

Population analysis: Population analysis was performed using NONMEM (Version 7.2, ICON, Hanover, MD) and the First Order Conditional Estimation method with interaction (FOCE INTER) was used to fit the model.[13] PsN version 3.4.2 and Xpose implemented in R was used in processing NONMEM 7 outputs.[14,15]

Model selection was guided by the plausibility and precision of parameter estimates, Akaike Information Criterion (AIC), as well as goodness-of-fit plots of observed concentrations versus time and conditional weighted residuals (CWRES) *versus* time.

Model development: Assuming that amlodipine concentrations are not influenced by simvastatin when the two drugs are co-administered, the model development was conducted by first developing a model for amlodipine and then developing a model for simvastatin and simvastatin acid, with the amlodipine model parameters being fixed at their *post hoc* estimates obtained at the previous step. In each step, the effects of CYP3A4/5 polymorphisms and covariates on the plasma concentrations of simvastatin, simvastatin acid and amlodipine were also explored. In developing the interaction model, simvastatin and simvastatin acid data were modelled simultaneously.

Model for amlodipine

The model for amlodipine was developed using amlodipine data only. Assuming first-order absorption, compartment models were tested up to a 3-compartment model, and influences of CYP3A4/5 polymorphisms and covariates were examined.

Model for simvastatin and simvastatin acid

Basic model: Simvastatin absorption kinetics were modeled by assessing various types of absorption processes including a single absorption process such as first-order absorption and Weibull absorption and a multiple absorption process such as simultaneous first- and zero-order absorption, sequential zero- and first-order absorption, and a series of transit compartment absorptions followed by first-order absorption.[16] Disposition kinetics of simvastatin and simvastatin acid were modeled by assessing a one-, a two- and a three-compartment model. For elimination kinetics, simvastatin clearance was assumed to be the sum of the hepatic clearance mediated by CYP3A4/5 and the conversion clearance from simvastatin to simvastatin acid mediated by esterase, paraoxonases, and non-enzymatic hydrolysis.

For inter-individual random effects, a proportional error model was used, and for intra-individual random effects, a combined error model was used to assess the relative contributions of additive and proportional error components. Random effect associated with crossover design was dealt with by including the inter-occasional variability (IOV) into the model.

Model for interaction with amlodipine: It was assumed that the interaction with amlodipine through CYP3A4/5 can occur in

both intestinal walls and hepatocytes. For the former, simvastatin bioavailability of co-administration was assumed to increase by a constant fraction due to the inhibited activity of CYP3A4/5 present on intestine walls, by the following relationship:

$$F_{SINGLE} = FR \times F_{CO} \quad (1)$$

where F_{SINGLE} and F_{CO} denote simvastatin bioavailability of single- and co-administrations, respectively, with $FR < 1$.

For the latter, the hepatic clearance of simvastatin and simvastatin acid was assumed to be inhibited by the decreased activity of CYP3A4/5 present in liver cells. In particular, the activity of CYP3A4/5 in liver cells was assumed to be inhibited associated with the plasma concentration of amlodipine as below:

$$INHCLP = CLP \times \left(1 - \frac{CP}{KI + CP}\right) \text{ for simvastatin} \quad (2a)$$

$$INHCLM = CLM \times \left(1 - \frac{CP}{KI + CP}\right) \text{ for simvastatin acid} \quad (2b)$$

where CLP and CLM are the clearances of the parent drug and the metabolite of simvastatin, $INHCLP$ and $INHCLM$ are their respective inhibited clearances, CP is amlodipine plasma concentration, and KI is the inhibition constant, representing the amlodipine concentration at 50% of maximum inhibition.

Model for influence by CYP polymorphisms: A mixture model was used as below to describe the potential polymorphism of CYP3A4/5 activities in simvastatin and simvastatin acid. This genetic polymorphism is expected to produce two subgroups of extensive (EM) and poor metabolizers (PM), with the different bioavailabilities due to different activity levels of CYP3A/5 enzymes present on the intestinal wall and with the different hepatic clearances due to different activity levels of CYP3A/5 enzymes present in liver cells. The difference in simvastatin bioavailability between the two subgroups was modeled as

$$BA_{EM} = \frac{\theta_{BA,EM} \times \exp(\eta_{BA,EM})}{1 + \theta_{BA,EM} \times \exp(\eta_{BA,EM})} \quad (3a)$$

$$BA_{PM} = 1 \quad (3b)$$

$$PRO_{EM} = \theta_{PRO,EM} \quad (3c)$$

$$PRO_{PM} = 1 - PRO_{EM} \quad (3d)$$

where $\theta_{BA,EM}$ and $\eta_{BA,EM}$ are fixed and random effects, respectively, for the bioavailability model for the EM subgroup with $0 < BA_{EM} < 1$, and $\mathrm{PRO_{EM}}$ is the proportion of the subjects belonging to the EM group. BA_{PM} was fixed to 1 to assume that the CYP3A4/5 activity in intestinal walls for the PM group was at its minimal level as compared to that for the EM group.

The difference in the hepatic clearance of the parent drug of simvastatin between the two subgroups was modeled as follows:

$$CLP_{EM} = \theta_{CLP,EM} \times \exp(\eta_{CLP}) \quad (4a)$$

$$CLP_{PM} = \theta_{CLP,PM} \times \exp(\eta_{CLP}) \quad (4b)$$

where $\theta_{CLP,EM}$ and $\theta_{CLP,PM}$ are fixed effects for the hepatic clearance model for the EM and PM subgroups, respectively, and η_{CLP} is the random effect.

The difference in the hepatic clearances of the metabolite of simvastatin between the two subgroups was similarly modeled as follows:

$$CLM_{EM} = \theta_{CLM,EM} \times \exp(\eta_{CLM}) \quad (4c)$$

$$CLM_{PM} = \theta_{CLM,PM} \times \exp(\eta_{CLM}) \quad (4d)$$

Model for covariate effects: As a final step in the model development, covariate analysis was undertaken to examine if there were any significant relationships between model parameters and the following demographic factors: age, weight, height, sex, smoking status, and alcohol consumption.

Stepwise covariate modeling (SCM) using PsN was performed at significance levels of $p < 0.05$ for forward selection and $p < 0.001$ for backward elimination. Here, due to the tremendous computational time required for covariate selection with the original simultaneous model developed, without loss of generality, the selection procedure was conducted separately for each sub-model in a sequential manner in the following order: sub-model for simvastatin for single-administration, sub-model for simvastatin acid for single-administration, sub-model for simvastatin for co-administration, and lastly the sub-model for simvastatin acid for co-administration.

Model evaluation: The final model was evaluated using a visual predictive check (VPC) given 1,000 datasets simulated from the final model. Specifically, the percent of observed concentrations falling outside the 90% prediction interval, defined by the range between the 5th and 95th percentiles, was assessed for each drug component.

Determination of an optimal simvastatin dose: Next, the typical parameter estimates obtained from the final model were used to predict an optimal simvastatin dose to minimize the increase in the steady-state concentrations of simvastatin and simvastatin acid for a co-administration with amlodipine, relative to a single-administration of simvastatin 40 mg only. The search for an optimal dose of simvastatin was made for a range of 2–50 mg, with a search interval of 2 mg. The optimal simvastatin dose D^* to be co-administered with amlodipine was defined as the dose that minimizes the absolute relative error (ERR, %) as given below, with $i = 1$ for simvastatin and $i = 2$ for simvastatin acid:

$$ERR = (ERR_1 + ERR_2)/2 \quad (5a)$$

$$ERR_i = \mathrm{abs}(C_{\max}C_i - C_{\max}S_i)/C_{\max}S_i \times 100, \ \ i = 1, \ 2 \quad (5b)$$

where $C_{max}C_i$ is the typical steady-state $C_{\max}$ of simvastatin ($i = 1$) or simvastatin acid ($i = 2$) predicted for a co-administration of simvastatin dose D and amlodipine 10 mg for nine days, and $C_{max}S_i$ is the typical steady-state $C_{\max}$ of simvastatin ($i = 1$) or simvastatin acid ($i = 2$) predicted for a single-administration of simvastatin 40 mg for nine days.

Results

Pharmacokinetic data: A total of 48 subjects were enrolled and 44 completed the study. Their demographic data, consisting of age, weight, and height as continuous covariates, and gender, smoking status and alcohol consumption as dichotomous covariates, are presented in **Table 1**.

According to preliminary analyses with a non-compartmental method, our data showed in the case of co-administrating simvastatin with amlodipine 1.8- and 1.9-fold increase for simvastatin and 1.9- and 2.3-fold increase for simvastatin acid in AUC and $C_{\max}$, respectively, as compared to a single administration of simvastatin (not shown). These preliminary results provided sufficient justification for using our data to develop an interaction model between the two drugs.

Model development:

Model for amlodipine

Amlodipine data were best described by a two-compartment

Table 1. Summary of demographic data for 48 healthy Korean male subjects

Continuous	Median	Range
Age (years)	24	22–50
Weight (kg)	69.85	52.9–87.1
Height (cm)	174.4	162–186.3
Categorical		
Gender (Male/Female)	48/0	—
Smoking (Yes/No)	29/19	—
Alcohol (Yes/No)	20/28	—
Xanthine (Yes/No)	19/29	—

model with first-order absorption. AIC values were 1,538 for the one-compartment model and 1,504 for the two-compartment model, with the estimation step not running successfully for a three-compartment model. The pharmacokinetic parameters for amlodipine estimated from the selected model are listed in **Table 2a**. Relative standard errors of model parameters were all below 30%. Age and weight were found to have a significant influence on clearance and peripheral volume of distribution ($p < 0.001$). Unlike the previous work,[7] no significant genetic difference was found when a mixture model was implemented. A goodness of fit for the selected model is drawn in **Figure 1a**, showing no trend in the distribution of residuals.

Model for simvastatin and simvastatin acid

Basic model: To select a model for absorption kinetics, simvastatin data from single-administration were analyzed preliminarily using a one-compartment model for disposition kinetics.

The resulting AIC values were 122.8 for first-order absorption, 89.8 for Weibull absorption, −45.2 for simultaneous first- and zero-order absorption, −43.2 for sequential zero- and first-order absorption, and 323.6 for a series of transit compartment absorptions followed by first-order absorption. Although Weibull and multiple absorption models gave smaller AIC values, first-order absorption was selected because the other types of absorption gave numerical difficulties in the rest of the model building process.

Next, to select a model for disposition kinetics given first-order absorption, simvastatin data were re-analyzed and found to be best described by a two-compartment model, yielding a smaller AIC value than a one-compartment model (129.3 *versus* 132.8). A three-compartment model was not taken into account because the estimation step did not run successfully.

Then, conditioned on absorption and disposition parameter estimates of simvastatin obtained at the previous steps, disposition kinetics for simvastatin acid were characterized and found to be best described by a one-compartment model. Here, because the metabolite clearance and its distribution volume were not separately identifiable unless the metabolite is administered alone or the metabolic fraction is known, the volume of distribution of the metabolite was fixed to that of the parent drug.[17]

Finally, simvastatin and simvastatin acid data were modeled simultaneously, based on the basic model for simvastatin and simvastatin acid consisting of a two-compartment model with first-order absorption for simvastatin, linked to a one-compartment model for simvastatin acid. In modelling random effects, correlations between model parameters were assumed to be zero because otherwise the estimation procedure did not run successfully.

Model for interaction with amlodipine: As described in the Methods section, when the relative bioavailability of simvastatin for single-administration was represented as a fraction of that for co-administration, it was estimated to be 0.68. This was equivalent to saying that the relative bioavailability of simvastatin for co-administration is about 1.46 times higher than that for single-administration.

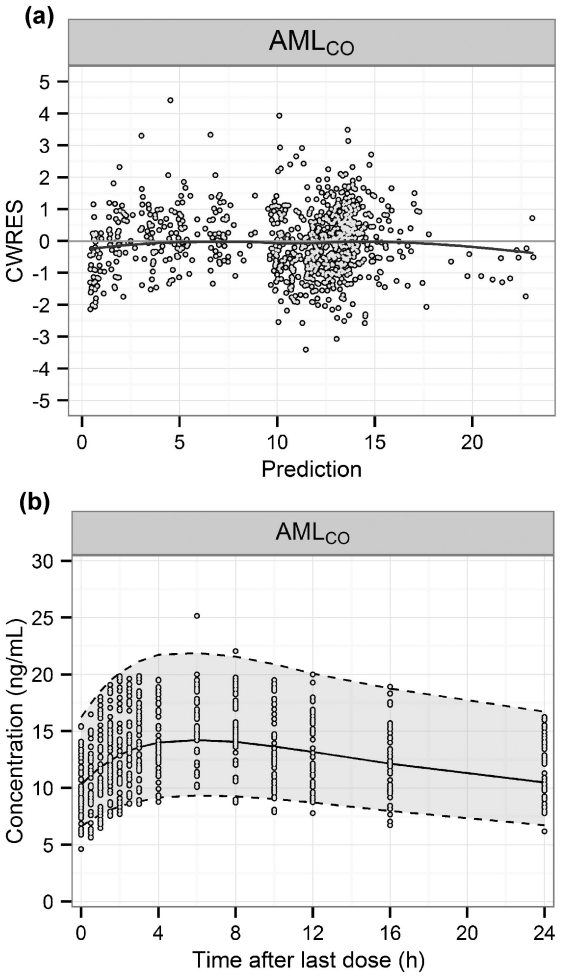

Fig. 1. Model evaluation for amlodipine
(a) A goodness of fit plot for the selected amlodipine model where AML_{CO} denotes the amlodipine concentration under co-administration (=simvastatin 40 mg and amlodipine 10 mg given together *q.d.* for 9 days), and CWRES and Prediction denote the conditional weighted residual and the model prediction, respectively. (b) Visual predictive check results for the amlodipine model obtained using 1,000 datasets simulated the final model, where dotted lines represent the 90% prediction interval and the solid line the median.

The influence of interaction with amlodipine on hepatic clearance of simvastatin and simvastatin acid was estimated to reach 50% of its maximum level at an amlodipine concentration of 92.8 ng/mL.

Model for influence by CYP polymorphisms: A mixture model to account for potential genetic variation of CYP3A5 activities in simvastatin metabolism was tested in three subgroups. Our results estimated the existence of two subgroups, with about 80% belonging to the EM and 20% to the PM group. The relative bioavailability of the EM group was estimated to be about 19% of that of the PM group.

As compared to the PM group, the CYP3A4/5 mediated hepatic clearance of the EM group was estimated to be 22% for simvastatin (130.5 *versus* 604.1 L/h), and 61% for simvastatin acid

Table 2. Parameter estimates of the final model

(a) Amlodipine (AML)

Parameter	Covariate model	θ (RSE%)	ω (CV %) (RSE%)	p-value
KA (h^{-1})	$KA = \theta_{KA}$	$\theta_{KA} = 0.2083$ (7.6%)		
V2 (L)	$V2 = \theta_{V2} \cdot \exp(\eta_{V2})$	$\theta_{V2} = 1{,}048$ (8.5%)	$\omega_{V2} = 32.7\%$ (22.7%)	
CL (L/h)	$CL = \theta_{CL} \cdot \{1 + \theta_{CL\ AGE} \cdot (AGE - 24.0)\} \cdot \exp(\eta_{CL})$	$\theta_{CL} = 32.38$ (3.6%)	$\omega_{CL} = 22.6\%$ (17.4%)	
		$\theta_{CL\ AGE} = -0.01995$ (10.1%)		< 0.001
V3(L)	$V3 = \theta_{V3} \cdot \{1 + \theta_{V3\ WT} \cdot (WT - 69.8)\}$	$\theta_{V3} = 969.7$ (7.3%)		
		$\theta_{V3\ WT} = 0.01587$ (31.4%)		< 0.001
Q (L/h)	$Q = \theta_{Q}$	$\theta_{Q} = 80.85$ (9.2%)		
Residual	$CP = \hat{CP} \cdot (1 + \varepsilon_{pro}) + \varepsilon_{add}$	$\sigma_{pro} = 11.7\%$ (11.0%)		
		$\sigma_{add} = 0.089$ (19.7%)		

KA, first-order absorption rate constant; V2, central volume of distribution; CL, hepatic clearance; V3, peripheral volume of distribution; Q, intercompartmental clearance; ω_{V2}, inter-individual variability of V2; ω_{CL}, inter-individual variability of CL; σ_{pro}, proportional component of residual variability; σ_{add}, additive component of residual variability; AGE, age (years); WT, weight (kg).
RSE, relative standard error (=standard error/estimated value).
ω_{V2}, ω_{CL}, and σ_{pro} are in the unit of coefficient of variation (CV).

(b) Simvastatin (SV) and simvastatin acid (SVA)

Parameter	Covariate model	θ (RSE%)	ω (CV %) (RSE%)
Simvastatin (SV)			
KA (h^{-1})	$KA = \theta_{KA} \cdot \exp(\eta_{KA})$	$\theta_{K12} = 0.3505$ (6.1%)	$\omega_{KA} = 30.1\%$ (34.1%)
V2 (L)	$V2 = \theta_{V2} \cdot \exp(\eta_{V2} + \kappa_{V2,j})$	$\theta_{V2} = 200.3$ (25.0%)	$\omega_{V2} = 29.7\%$ (75.9%)
			$\pi_{V2} = 45.8\%$ (29.1%)
CLP (L/h)	$CLP = \theta_{CLP} \cdot \exp(\eta_{CLP})$	$\theta_{CLP(EM)} = 130.5$ (27.7%)	$\omega_{CLP} = 62.3\%$ (43.8%)
		$\theta_{CLP(PM)} = 604.1$ (26.4%)	
CLC (L/h)	$CLC = \theta_{CLC}$	$\theta_{CLC} = 123.8$ (26.2%)	
V3 (L)	$V3 = \theta_{V3}$	$\theta_{V3} = 2{,}322$ (26.0%)	
Q (L/h)	$Q = \theta_{Q} \cdot \exp(\kappa_{Q,j})$	$\theta_{Q} = 62.06$ (26.0%)	$\pi_{Q} = 155.2\%$ (32.1%)
F	$F_{SINGLE} = FR \cdot F_{CO}$		
	$F_{CO} = BA$	$\theta_{BA,E} = 0.2334$ (17.3%)	
BA	$BA_{EM} = \theta_{BA,EM} \cdot \exp(\eta_{BA,EM}) / \{1 + \theta_{BA,EM} \cdot \exp(\eta_{BA,EM})\}$		$\omega_{BA,EM} = 44.9\%$ (35.9%)
	$BA_{PM} = 1$	$\theta_{FR} = 2.152$ (24.9%)	
FR	$FR = \theta_{FR} / (1 + \theta_{FR})$		
KI (ng/mL)	$KI = \theta_{KI} \cdot \exp(\eta_{KI})$	$\theta_{KI} = 92.83$ (32.2%)	$\omega_{KI} = 159.8\%$ (28.0%)
PRO	$PRO_{EM} = \theta_{PRO,EM}$	$\theta_{PRO,EM} = 0.7965$ (10.1%)	
	$PRO_{PM} = 1 - PRO_{EM}$		
Residual	$CP_{SV} = \hat{CP}_{SV} \cdot (1 + \varepsilon_{SV,pro}) + \varepsilon_{SV,add}$	$\sigma_{SV,pro} = 52.8\%$ (7.9%)	
		$\sigma_{SV,add} = 0.0035$ (23.6%)	
Simvastatin acid (SVA)			
CLM (L/h)	$CLM = \theta_{CLM} \cdot \exp(\eta_{CLM})$	$\theta_{CLM(EM)} = 73.06$ (25.3%)	$\omega_{CLM} = 38.5\%$ (32.7%)
		$\theta_{CLM(PM)} = 120.6$ (24.9%)	
V4 (L)	$V4 = V2$		
Residual	$CP_{SVA} = \hat{CP}_{SVA} \cdot (1 + \varepsilon_{SVA,pro}) + \varepsilon_{SVA,add}$	$\sigma_{SVA,pro} = 36.8\%$ (12.6%)	
		$\sigma_{SVA,add} = 0.012$ (50.6%)	

KA, first-order absorption rate constant; V2, central volume of distribution of parent drug; V3, peripheral volume of distribution of parent drug; CLP, hepatic clearance of parent drug; CLC, conversion clearance of parent drug; Q, inter-compartmental clearance of parent drug; F, bioavailability difference with formulation; BA, bioavailability difference with subgroup; FR, bioavailability ratio of single- to coadministration; KI, inhibition constant; PRO, proportion of the subjects belonging to each subgroup; CLM, hepatic clearance of metabolite; V4, volume of distribution of metabolite; ω_{KA}, inter-individual variability of KA; ω_{V2}, inter-individual variability of V2; ω_{CLP}, inter-individual variability of CLP; ω_{BA}, inter-individual variability of BA; ω_{KI}, inter-individual variability of KI; ω_{CLM}, inter-individual variability of CLM; $\kappa_{V2,j}$, inter-occasional random effect of V2 for single- ($j = 1$) and co-administration ($j = 2$); π_{V2}, variability of $\kappa_{V2,j}$; $\kappa_{Q,j}$, inter-occasional random effect of Q for single- ($j = 1$) and co-administration ($j = 2$); π_{Q}, variability of $\kappa_{Q,j}$; σ_{pro}, proportional component of residual variability; σ_{add}, additive component of residual variability.
SINGLE, single-administration (=simvastatin administered only); CO, co-administration (=simvastatin and amlodipine administered concomitantly); PM, poor metabolizers; EM, extensive metabolizers.

(73.06 *versus* 120.6 L/h), equivalent to saying that the CYP3A4/5 mediated hepatic clearance of the PM group is about 4.63 and 1.65 times higher than that of simvastatin and simvastatin acid, respectively.

Model for covariate effects: For covariate selection, no covariate was found to be significant. Although age was found to have a significant effect on clearance, it did not reach the significance level of $p < 0.001$ in the backward deletion and was excluded.

Parameter estimates for the final model for simvastatin and simvastatin acid are listed in **Table 2b**. The coefficient of variation for random effects was >50% in the inter-individual variability of CLP and KI, and the inter-occasional variability of Q. For KA, V3, and Q of amlodipine, the inter-individual random effect was not considered because they gave relative standard errors larger than 50%. For CLC, the inter-individual random effect was not included in the model because the run did not converge when it was included. CWRES *versus* prediction plots are given in **Figure 2a**, indicating a good agreement between the final model and the data. The final model structure for simvastatin, simvastatin acid and amlodipine is illustrated in **Figure 3**.

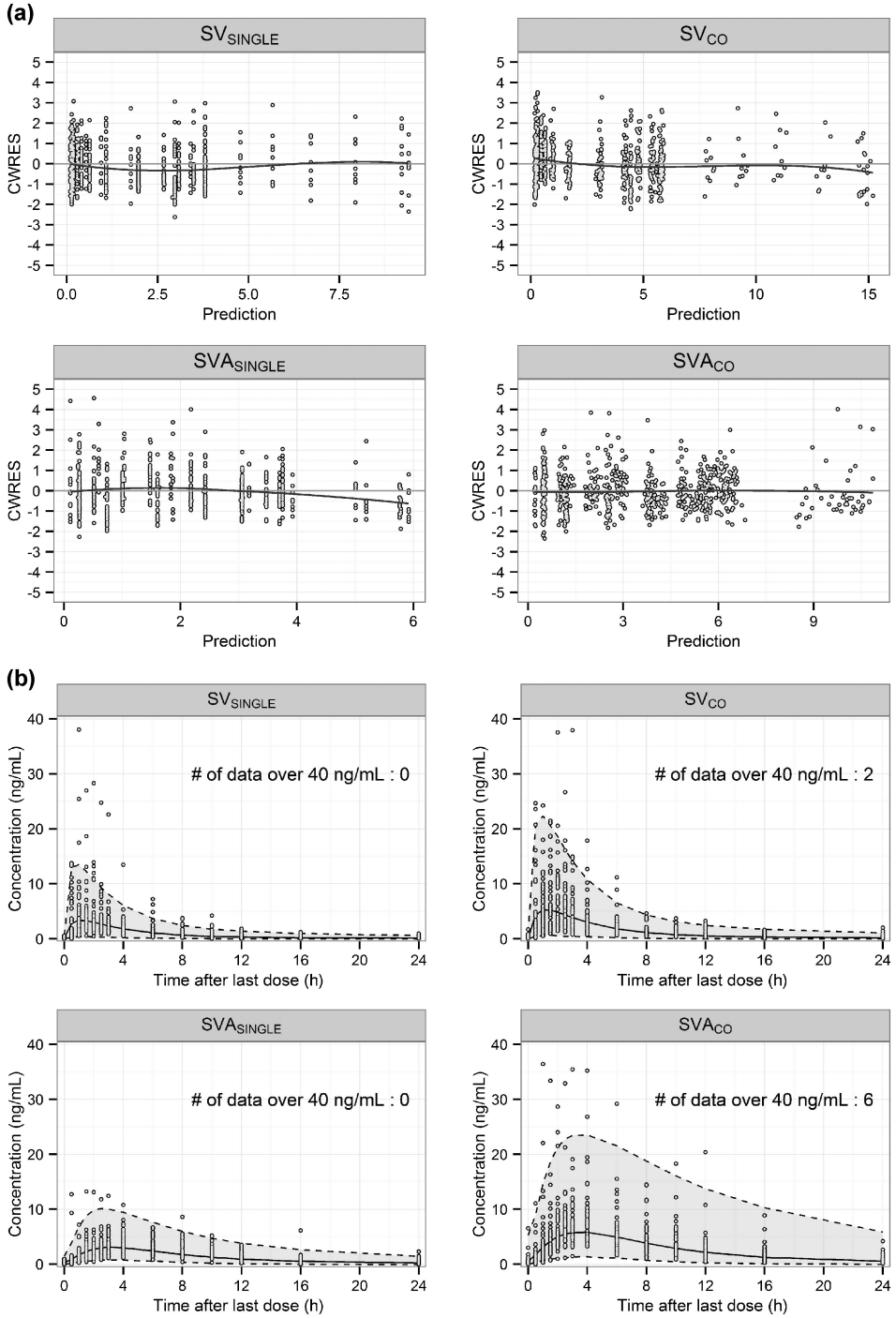

Fig. 2. Model evaluation for simvastatin and simvastatin acid
(a) Goodness of fit plots for the selected model for simvastatin and simvastatin acid concentration where SV_{SINGLE} and SVA_{SINGLE} denote simvastatin and simvastatin acid for single-administration (=simvastatin 40 mg given alone *q.d.* for 9 days), respectively, SV_{CO} and SVA_{CO} denote simvastatin and simvastatin acid for co-administration (=simvastatin 40 mg and amlodipine 10 mg given together *q.d.* for 9 days), respectively, and CWRES and Prediction denote the conditional weighted residual and the model prediction, respectively. (b) Visual predictive check results for the model for simvastatin and simvastatin acid obtained using 1,000 datasets simulated from the final model, where dotted lines represent the 90% prediction interval and the solid line the median.

Model evaluation: VPC results obtained from 1,000 datasets simulating the final model are depicted in **Figure 1b** for amlodipine and **Figure 2b** for simvastatin and simvastatin acid. For the model for amlodipine, 6.4% of observations fell outside the 90% prediction interval, and for the model for simvastatin and simvastatin acid, 4.4% and 1.8% of observations for co-administration and 1.0% and 1.3% of observations for single-administration fell outside their respective 90% prediction interval,

indicating the adequacy of the developed model.

Determination of an optimal simvastatin dose: The dose optimization results using the typical parameter estimates obtained from the final model are summarized in **Table 3** and the predicted concentrations of simvastatin and simvastatin acid given the optimal dose are shown in **Figure 4**. The optimal simvastatin dose D^* that minimizes the concentration increase when co-administered *q.d.* with amlodipine 10 mg was estimated to be 24 mg for both the PM and EM groups. Note in the table that $C_{max}S$ becomes higher in the EM group.

Table 3. The optimal dosage regimen results for simvastatin that minimize the interaction with amlodipine in co-administration

	$C_{max}S$ (ng/mL)	Dose (mg)	$C_{max}C_1$ (ng/mL)	$C_{max}C_2$ (ng/mL)	ERR_1 (%)	ERR_2 (%)	ERR (%)
EM	$C_{max}S_1 = 3.81$	24	3.49	3.77	8.4	0.5	4.4
	$C_{max}S_2 = 3.79$	40	5.81	6.28	52.7	65.9	59.3
PM	$C_{max}S_1 = 9.41$	24	8.82	6.13	6.3	3.6	4.9
	$C_{max}S_2 = 5.92$	40	14.70	10.22	56.2	72.6	64.4

The result for a simvastatin dose of 40 mg was listed together for comparison. For symbols, see text.

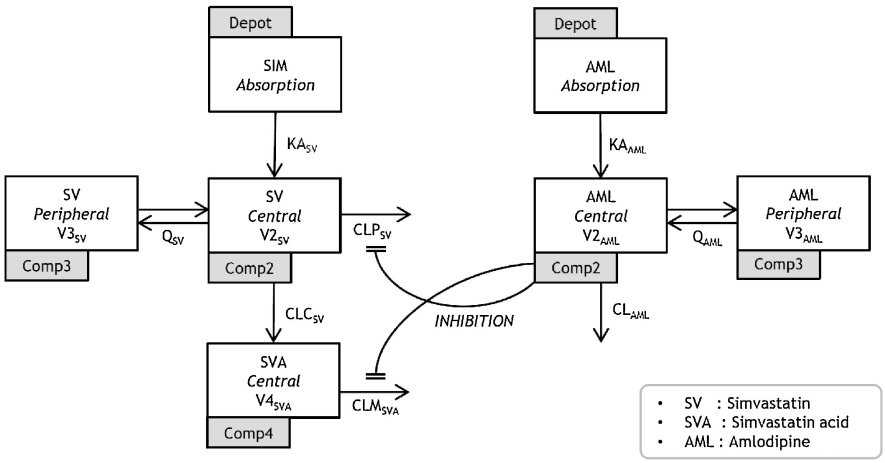

Fig. 3. The final model structure for simvastatin, simvastatin acid and amlodipine

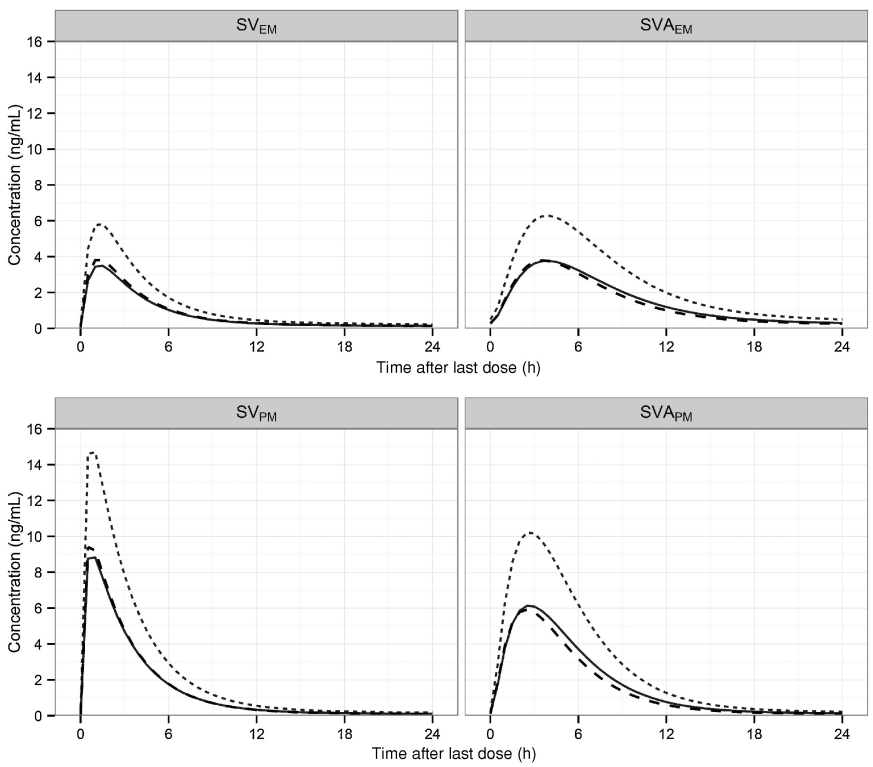

Fig. 4. Concentrations of simvastatin (SV) (left panels) and simvastatin acid (SVA) (right panels) simulated from the final model for a daily dose of simvastatin 24 mg co-administered with amlodipine 10 mg for nine days (solid line), superimposed on predicted concentrations for single-administration of simvastatin 40 mg only (dashed line), obtained in EM (upper panels) and PM groups (lower panels)
Predicted concentrations for co-administration of simvastatin 40 mg with amlodipine 10 mg were also plotted for comparison (dotted line).

Discussion

In this study, we developed a model-based approach to characterize the drug-drug interaction between simvastatin and amlodipine and how it can be used in selecting an optimal simvastatin dose that can avoid an unwanted increase in simvastatin plasma concentration, thereby minimizing the risk of adverse drug reactions while maintaining drug efficacy.

The selected interaction model was described using a two-compartment model with first-order absorption for simvastatin, linked to a one-compartment model for simvastatin acid, given a two-compartment model with first-order absorption for amlodipine.

The important model components characterizing the drug-drug interaction between simvastatin and amlodipine were (1) the inhibitory effect of amlodipine on simvastatin metabolism modeled as a function of amlodipine plasma concentration; and (2) CYP3A4/5 polymorphisms in simvastatin bioavailability and hepatic clearance implemented by the mixture model.

Simvastatin has been reported to have a moderate binding affinity to CYP3A4/5 and might be more easily detached by other substrates with a higher binding affinity such as amlodipine. Thus, during the modeling process, we assumed that amlodipine competitively inhibited the metabolism of simvastatin and simvastatin acid, but not vice versa. This was experimentally confirmed in our separate trial where for the same subjects who participated in this study amlodipine 10 mg was given alone *q.d.*; resulting AUC and C_{max} of amlodipine were found to be not significantly different from those for co-administration (not shown). Thus, the amlodipine data were modeled separately and the *post hoc* parameter estimates of amlodipine were used in the parent-metabolite model of simvastatin in a sequential manner.

Given the reversible interconversion between simvastatin and simvastatin acid, we initially tried including in the model the rate constants of both the forward conversion of simvastatin to simvastatin acid and the backward conversion of simvastatin acid to simvastatin. However, the model did not successfully converge when the two conversion rate constants were included because there was strong correlation between the parent and metabolite concentrations and the two conversion rate constants could not be estimated separately.

For the inhibitory effect of amlodipine on simvastatin metabolism, the simvastatin bioavailability for co-administration increased 1.46-fold, compared to that of single-administration. This increase was associated with reduced activity of CYP3A4/5 located on the intestine, which is a major site responsible for low oral bioavailability when the activity of CYP enzymes is inhibited.[18] In our analysis, the inhibition rate constant *KI*, which was estimated to be 92.8 ng/mL, was modeled to be common to simvastatin and simvastatin acid because it was legitimate to assume that the inhibition mechanism by amlodipine concentration would not differ between the parent drug and metabolite. This means a 13% decrease in the clearance of both simvastatin and simvastatin acid when evaluated at the typical C_{max} of amlodipine, which was predicted to be 14 ng/mL for the daily amlodipine dose of 10 mg.

In the mixture model, the metabolic clearance of simvastatin and simvastatin acid was allowed to vary between the EM and PM groups because both moieties are metabolized by CYP3A4/5, which has potential genetic polymorphisms. Interestingly, as reported in **Table 2**, the result was that the hepatic clearance of the EM subgroup was lower than that of the PM subgroup in both simvastatin and simvastatin acid. The reason would be that because for simvastatin the first-pass metabolism takes place largely in the gut wall, the amount of simvastatin entering into the systemic circulation becomes much lower in the EM group than in the PM group, leading to the reduced amount of simvastatin available in hepatic metabolism in the EM group. As a result, the hepatic clearance of simvastatin in the EM group could become lower than in the PM group. Further, as the blood amount of simvastatin acid would be smaller in the EM group due to the smaller amount of simvastatin available for its conversion to simvastatin acid, the resulting hepatic clearance of simvastatin acid in the EM group could become lower than in the PM group. This was also confirmed by a noncompartmental analysis of the terminal half-life, which was found to be longer in the EM group (not shown), supporting the lower clearance of this subgroup obtained from the modeling result. **Table 2** also reports that the between-group difference in simvastatin acid clearance was smaller than for simvastatin clearance (1.65 times *versus* 4.63 times). A possible explanation for this is that the metabolite is a byproduct of the metabolism of the parent drug, and a large portion of between-group differences in simvastatin acid concentration was inherited from the between-group difference in simvastatin concentration, and thus only a small portion is accounted for by the additional difference in simvastatin acid concentration. In modeling the inter-individual variability, we assumed it was the same for the 2 subgroups [Eq. (4)], because the estimation step did not run successfully when it was allowed to vary.

Elevated simvastatin concentrations occurring from drug-drug interactions can cause adverse reactions such as rhabdomyolysis, which can be a serious problem, particularly in the PM group. Such drug interactions can be avoided by adjusting the dosage and/or dosing time.[19] According to **Table 2a**, however, amlodipine half-life was estimated to be as long as 22 h, indicating adjusting dosing time would be almost unrealistic. Therefore, as an alternative, adjusting the simvastatin dose was suggested in this work based on the model developed. Our results showed that, in both the EM and PM groups, simvastatin 24 mg was an optimal dose in a co-administration with amlodipine 10 mg that can minimize the concentration increase in simvastatin and simvastatin acid due to drug-drug interaction with amlodipine, thereby achieving a concentration profile similar to that for single-administration of simvastatin 40 mg only. In other words, when simvastatin 40 mg is co-administered with amlodipine 10 mg, we suggest that a 40% dose reduction of simvastatin is needed to avoid drug-drug interactions.

Although no covariate was found to be significant in our study, age has been reported to be influential on clearance in studies with larger populations.[20]

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