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Article

Prediction of the Impact of *CYP2C19* Polymorphism on Drug-Drug Interaction between Voriconazole and Tacrolimus Using Physiologically-Based Pharmacokinetic Modelling

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Voriconazole increases tacrolimus blood concentration significantly when coadministrated. The recommendation of reducing tacrolimus to 1/3 in voriconazole package insert seems not to be satisfactory in clinical practice. In vitro studies demonstrated that the magnitude of inhibition depends on the concentration of voriconazole, while voriconazole exposure is determined by the genotype status of *CYP2C19. CYP2C19* gene polymorphism challenges the management of drug-drug interactions(DDIs) between voriconazole and tacrolimus. This work aimed to predict the impact of *CYP2C19* polymorphism on the DDIs by using physiologically based pharmacokinetics (PBPK) models. The precision of the developed voriconazole and tacrolimus models was reasonable by evaluating the pharmacokinetic parameters fold error, such as AUC_{0-24}, C_{max} and t_{max}. Voriconazole increased tacrolimus concentration immediately in all population. The simulated duration of DDIs disappearance after voriconazole withdrawal were 146h, 90h and 66h in poor metabolizers (PMs), intermediate metabolizers (IMs) and extensive metabolizers(EMs), respectively. The developed and optimized PBPK models in this study can be applied to assit the dose adjustment for tacrolimus with and without voriconazole.

Keywords: Voriconazole. Tacrolimus. *CYP2C19* gene polymorphism. Physiologically based pharmacokinetics (PBPK) model. Drug-drug interaction.

INTRODUCTION

Voriconazole is a second-generation triazole antifungal agent with potent activity against a broad spectrum of pathogens. It has been approved by the Food and Drug Administration (FDA) for "the treatment of invasive aspergillosis, esophageal candidiasis, candidemia in non-neutropenic patients, disseminated Candida infections, and infections caused by Scedosporium apiospermum and Fusarium spp (Pfizer, 2014)".Voriconazole is primarily metabolized by hepatic cytochrome P450(*CYP*) enzymes *2C19* and *3A4*, with minimal involvement of *CYP2C9* (Hyland, Jones, Smith, 2003), and demonstrates saturable, nonlinear pharmacokinetics in adults (Purkins *et al.*, 2002). Besides, voriconazole is known as the inhibitor of *CYP3A* (*CYP3A4/5*) enzymes, which indicates the drugs metabolized by *CYP3A* may be influenced by voriconazole (Dresser, Spence, Bailey, 2000; Mori *et al.*, 2012).

Tacrolimus, an immunosuppressive agent to prevent or treat allograft rejection, is commonly used in solid organ

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transplant (SOT) patients due to the high risk of invasive aspergillosis(IA) (Husain, Camargo, 2019). However, tacrolimus has a narrow therapeutic blood concentration range. Overexposure increases the therapeutic effect while enhancing the risk the toxicity and infection. On the other side, low drug blood concentration may lead to a higher risk of graft rejection. As tacrolimus is the substrate of *CYP3A*, concomitant administration of voriconazole and tacrolimus results in a significant increase in the latter's blood concentration. Hence dose adjustment of tacrolimus is typically required in clinical practice.

Although the voriconazole package insert recommends reducing tacrolimus to 1/3 and carrying out therapeutic drug monitoring(TDM), the magnitude of the interaction is highly variable and the empiric dose reduction seems not be satisfactory. In vitro studies demonstrated that the magnitude of inhibition of tacrolimus metabolism by *CYP3A* depends on the concentration of voriconazole (Venkataramanan *et al*., 2002; S. Zhang *et al*., 2012), while voriconazole exposure is affected by CYP2C19 gene polymorphism. Clinical studies illustrated that the voriconazole $AUC_{0-\infty}$ was 2.8 to 4.1 times higher in poor metabolizers (PMs) compared to extensive metabolizers (EMs) (Moriyama *et al*., 2015). The voriconazole package insert noted that in healthy Caucasians and Japanese, voriconazole exposure of PMs and intermediate metabolizers(IMs)was 4 and 2 times higher than EMs, respectively. Besides, when voriconazole is discontinued, the duration of its impact on tacrolimus is uncertain. Worth mentioning that the goal trough concentration range of tacrolimus varies with transplanted organs and the time after transplantation (Staatz, Tett, 2004). Therefore, the management of drug-drug interactions (DDIs) remains challenging.

Physiologically based pharmacokinetics (PBPK) models can integrate in vitro ADME (absorption, distribution, metabolism and excretion), in vivo PK and interaction information, which is widely used in pharmaceutical research. In the past decade, it has consistant growth in the number of new drug applications submitted to the FDA that contained PBPK analyses. It was reported that about 60% of the expected purpose of PBPK analyses including in these submissions was mainly to assess enzyme-based DDIs (Grimstein *et al*., 2019). The reliability of DDIs prediction function of well-designed PBPK models has been approved by the U.S. FDA and European Medicines Agency (EMA) (Access October 10, 2019; Drug interaction studies-study design, Access October 10, 2019). Theoretically, PBPK models can accurately assess the magnitude of the impact of *CYP2C19* gene polymorphism on DDIs between voriconazole and tacrolimus, which can be reliable tools to provide individualized dose adjustment of tacrolimus.

MATERIAL AND METHODS

In this study, Simcyp® (version 16, Simcyp, Sheffield, UK) was utilized for PBPK model establishment. Three models for voriconazole of different metabolism gene populations were modified and refined based on the Bharat Damle, et al. (Damle, Varma, Wood, 2011). The initial tacrolimus PBPK model was optimized(H. Zhang *et al*., 2018) and verified all PBPK models performance using observed data in the literature to predict DDIs between voriconazole and tacrolimus. After reasonably predicted the DDIs between voriconazole and tactrolimus when concomitant, the duration of serum concentration of tacrolimus returned to baseline after discontinuing voriconazole was also simulated. All models were carried out in healthy Chinese volunteers from 18 to 65. The workflow of the study is presented in Figure 1.

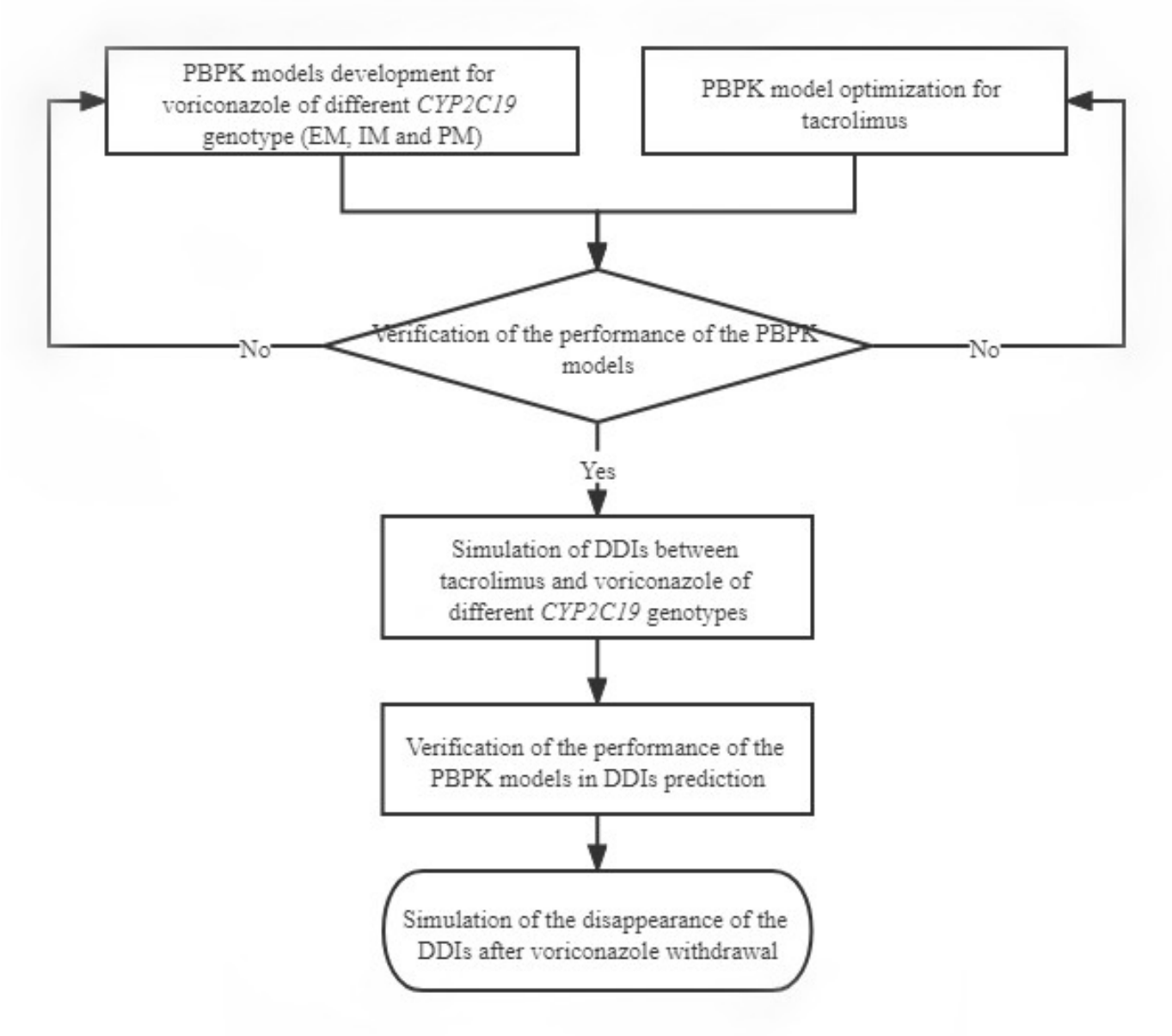

FIGURE 1 - The workflow of the study.

PBPK models development for voriconazole

Definition of three types of metabolizer

There are over thirty identified *CYP2C19* alleles with significant ethnic differences(Access March 30, 2021). The allele (*1) has a full function associated with regular *CYP2C19* activity. *CYP2C19*2* and *CYP2C19*3* are the two most common nonfunctional alleles, besides *CPY2C19*4*, *CYP2C19*5*, *CYP2C19*6* and *CYP2C19*8* (Zhou, Ingelman-Sundberg, Lauschke, 2017). Individuals with homozygous for *CYP2C19*1* are defined as EMs while those with two null alleles are defined as PMs. IMs carry one nonfunctional and one functional allele.

PBPK models development for voriconazole of different CYP2C19 genotype

To obtain the physicochemical parameters, in vitro data and clinical pharmacokinetic parameters

of voriconazole for PBPK models development, extensive literature research was carried out. The drug-specific components and parameters to develop the initial PBPK model were mainly from the published literatures (Chan *et al.*, 2013; Damle, Varma, Wood, 2011). However, the initial model did not take different *CYP2C19* genotypes into consideration, which may overestimate or underestimate the in vivo exposure of voriconazole and affect clinical outcomes. Therefore, the initial model was subdivided into three models using the observed data from different *CYP2C19* genotype populations and the modeling parameters were listed in Table I. The first-order model and minimal PBPK model were selected to describe the absorption module and distribution module of voriconazole, respectively. Given that *CYP2C19* has minimal effect on voriconazole metabolism in PMs, blocking this enzyme in the module can help the concentration-time curve match the observed data. Also, a more reasonable value of kinetic parameter V_{max} (maximum velocity) of CYPs (except *2C19*) can be obtained, which approximately equals to *CYP3A4*'s V_{max}. Then, the V_{max} value of *CYP2C19* was obtained by making the optimized model's concentration-time curve match the EMs' observed data best. Halving the *CPY2C19* enzyme abundance in Simcyp® library, the observed data of IMs was used to verify the performance of the optimized parameters and models. All trials of the simulation were conducted with a virtual population of 100 (10 trials of 10 subjects per trial) healthy Chinese volunteers (fasted state). Mean plasma profile data from literature were digitized using GetData Graph Digitizer (version 2.2)

The accuracy of prediction was assessed by the fold error(FE)[Eq.1] between the simulated and observed C_{max} or AUC. The FE value less than 2 indicated the results of the simulated value matched the observed value well.

$$\text{Fold error} = \frac{simulated\ value}{observed\ value} \text{ (if simulated>observed)}$$

or Eq.1

$$\text{Fold error} = \frac{observed\ value}{simulated\ value} \text{ (if observed> simulated)}$$

PBPK model optimization for tacrolimus

Most physicochemical and ADME parameters of tacrolimus for initial model development were obtained from our former study (H. Zhang *et al.*, 2018). In this study, pharmacokinetic parameters, such as blood to plasma (B/P), adipose, k_a (absorption rate constant), f_a (fraction absorbed) and V_{ss} (volume of distribution at steady state), were optimized to improve the initial model's performance, especially for the time of reaching a steady state. The modeling parameters were listed in Table-S1. The FE value was used to evaluate the precision of the optimized model.

Simulations of DDIs between Tacrolimus and Voriconazole of different *CYP2C19* genotypes

Simulations of the DDIs when concomitant administration

Both voriconazole and tacrolimus PBPK models were validated with published clinical study data (Imamura *et al.*, 2016). To assess the accuracy of prediction, these two models were utilized to simulate the data of DDIs between tacrolimus and voriconazole of different genotypes. With the same tacrolimus dose, voriconazole 400 mg was given every 12 hours on day 1, followed by 200 mg every 12 hours on day 2,3 and a single 200 mg dose in the morning on day 4. The DDIs were simulated in 100 virtual subjects by Simcyp®. The precision of DDI prediction was evaluated by FE, and the FE value within 2 indicated that the models could describe the DDIs well. For further verification of the predictive performance of the PBPK models, the ratio of model-predicted mean exposure changes of tacrolimus (area under the concentration-time curve ratio, AUCR) to observed values ($R_{predicted/observed}$) was calculated [Eq.2]. $R_{predicted/observed}$ value between 0.5-2 indicated that the predictive accuracy of PBPK modeling was reasonable (Vieira *et al.*, 2014).

$$R_{predicted/observed} = \frac{Model-predicted\ AUCR}{Clinical\ observed\ AUCR} \quad \text{Eq.2}$$

Simulations of the disappearance of the DDIs after voriconazole withdrawal

In order to simulate the time interval of voriconazole-tacrolimus interaction after voriconazole discontinuation in different genotype populations, a clinical scene in a virtual population of 100 (10 trials of 10 subjects per trial) was set according to the following protocol (1) give tacrolimus 1.5 mg po q12h as initial regimen; (2)input voriconazole 200mg po bid after reaching the steady concentration; (3) withdrawal voriconazole after tacrolimus reaching the new steady concentration; (4) count the time that tacrolimus AUC_{0-12} decreased to baseline as the duration of DDI disappearance; (5) repeat the steps above in different genotype populations.

RESULTS

PBPK models verification for voriconazole according to different *CYP2C19* genotype

The initial model was developed with the pharmacokinetic data from oral administration of voriconazole, without considering *CYP2C19* polymorphism in the metabolism period. Hence, the modified model parameters were optimized for the elimination parameter, V_{max}, by using the clinical studies data. According to our simulation, when the V_{max} value of *CYP2C19* and *CYP3A4* reached 3 pmol/min/pmol and 0.21 pmol/min/pmol, respectively (Table I), the simulated concentration-time curve fit the observed data best (Figure 2). The predicted pharmacokinetic parameters such as AUC_{0-12}, C_{max} and T_{max} of voriconazole according to different *CYP2C19* genotypes were all within 2-fold error compared with the observed data (Imamura *et al.*, 2016) (Table II). The predicted AUC_{0-12} of PMs and IMs was 3.51 and 1.54 times higher than EMs, respectively. The predicted C_{max} of PMs and IMs was 2.66 and 1.37 times higher than EMs, respectively.

TABLE I - Parameters for voriconazole PBPK models of different CYP2C19 genotype

Parameters	**Input Valuea**		
Physicochemical properties			
MW(g/mol)	349		
log $P_{o:w}$	1.8		
Compound type	Monoprotic base		
pK_a	1.76		
B/P	1.229		
Fraction unbound	0.42		
First order absorption model			
f_a	0.96		
$k_a(h^{-1})$	1.44		
$f_{u,gut}$	1		
$P_{app,caco-2}(10^{-6}cm/s)$	28.10		
Minimal PBPK distribution model			
V_{ss}(L/kg)	1.079		
Liver K_p	1		
Elimination			
In *vitro* metabolic system Recombinant			
Pathway	Pathway 1	Pathway 1	Pathway 2
Enzyme	*CYP2C19*	*CYP3A4*	*CYP3A4*
V_{max}(pmol/min/pmol)	3[b]	0.21[b]	0.10
K_m(μmol/L)	3.5	15	11

MW, molecular weight; log P, n-octanol:buffer partition coefficient; pK_a, acid dissociation constant; B/P, blood to plasma ratio; f_a, fraction absorbed; k_a, absorption rate constant; $f_{u,gut}$, unbounded compound fraction of gut; $P_{app,caco-2}$, Caco-2 cell permeation; V_{ss}, volume of distribution at steady state; V_{max}, maximum reaction velocity; K_m, Michaelis-Menten constant.

[a] The input values were main from reference (Damle, Varma, *et al.*, 2011).

[b] The values were optimized as explained in material and methods session.

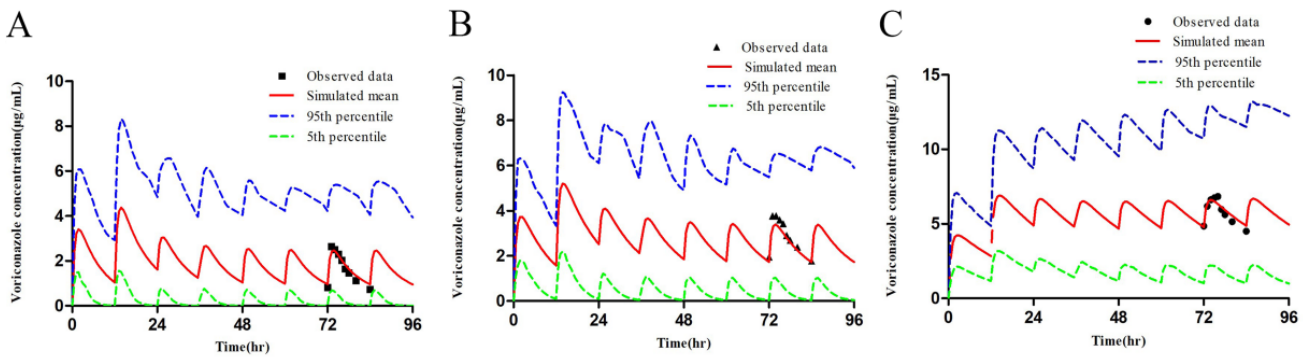

FIGURE 2 - Simulation of voriconazole plasma concentration-time profiles at steady state (200mg twice daily orally) in CPY2C19 EMs (A), IMs (B) and PMs (C). Solid line represents the mean value of the simulated population. Dash line represent the 5th-95th percentiles of simulated population. Green, red and blue circles represent the observed data for EMs, IMs and PMs, respectively.

TABLE II – The predicted versus observed pharmacokinetic parameters of voriconazole according to CYP2C19 genotype

CYP2C19 Genotype	AUC0-12(μg·h/mL)			Cmax(μg/mL)			Tmax(hr)		
	Predicted	Observed	Fold error	Predicted	Observed	Fold error	Predicted	Observed	Fold error
EM	19.88	18.8	1.06	2.49	2.8	1.12	1.74	1.5	1.16
IM	30.66	33.6	1.10	3.41	4.0	1.17	1.87	1.5	1.25
PM	69.8	67.8	1.03	6.63	7	1.06	2.07	2.8	1.35

Tacrolimus PBPK model verification

In this study, the full tacrolimus PBPK model prediction method 1 developed by Poulin and Theil (Poulin, Theil, 2002) was used to optimize the initial model. Pharmacokinetic parameters, such as f_a (0.968) and k_a (1.331 h^{-1}) in the absorption module, V_{ss} (3.311 L/kg) in distribution, were predicted by Simcyp. Meanwhile, setting the adipose value as 50 and blood to plasma value as 10 in the distribution module, the simulated concentration-time curve fit the observed data well (Figure S1). The simulated tacrolimus pharmacokinetic profiles (AUC_{0-24}, C_{max} and T_{max}) after a single oral dose (3mg) were compared with the observed data, and the results were all within twofold error (Table III).

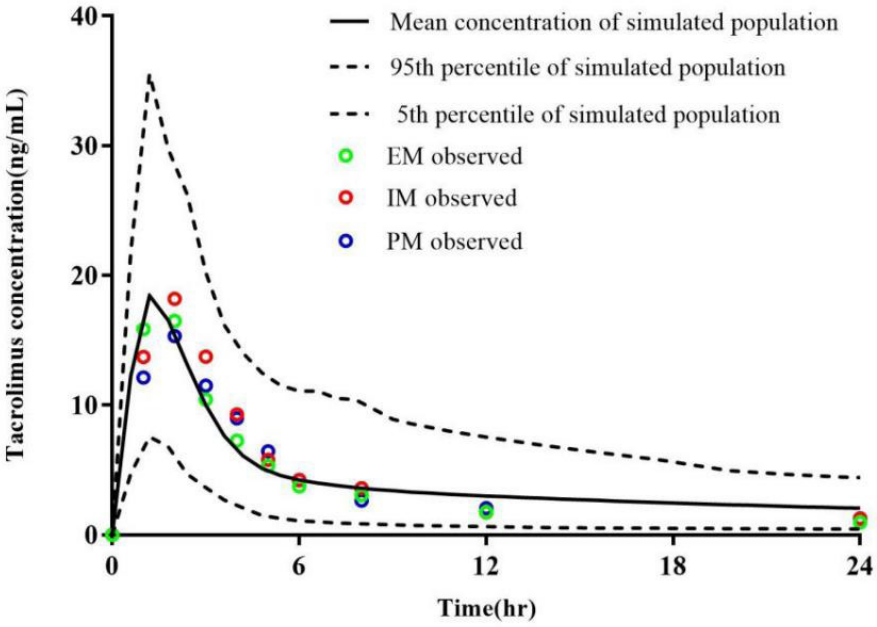

FIGURE S1 - Simulation of tacrolimus whole-blood concentration-time profiles after a 3mg single oral dose alone. Solid line represents the mean value of the simulated population. Dash lines represent the 5th and 95th percentiles value of the simulated population. Green, red and blue circles represent the observed data for EMs, IMs and PMs, respectively.

TABLE III - The predicted versus observed pharmacokinetic parameters of tacrolimus according to CYP2C19 genotype

	Parameters						
		***CYP2C19* Genotype**					
	predicted	**EM**		**IM**		**PM**	
		observed	**FE**	**observed**	**FE**	**observed**	**FE**
AUC_{0-24}(ng·h/mL)	108.239	88.3	1.23	108.2	1	94.8	1.14
C_{max}(ng/mL)	18.8667	18.3	1.3	20.5	1.09	16.3	1.16
T_{max}(h)	1.41	1.5	1.06	1.7	1.21	1.8	1.28

DDIs simulations between tacrolimus and voriconazole

Simulation of the DDIs when concomitant use of tacrolimus and voriconazole

The verified PBPK models with the incorporation of in vitro *CYP3A4*K_i (inhibition constant) for voriconazole (0.66μM) (Jeong, Nguyen, Desta, 2009) were used to simulate the effect of voriconazole on tacrolimus pharmacokinetics. After reaching the steady state with a single oral dose (3 mg), voriconazole 200mg twice daily was input, and the simulated tacrolimus whole-blood concentration-time curve of different *CYP2C19* genotype fit the observed data well (Figure 3). Compared with the observed data, the fold errors of C_{max} and AUC_{0-12h} were all less than 2 (Table IV). Furthermore, the $R_{predicted/observed}$ of AUCR was 0.69, 0.66, and 0.64 for EMs, IMs and PMs, respectively. The results indicated that the developed PBPK models can reasonably predict the DDIs between tacrolimus and voriconazole with different *CPY2C19* genotypes.

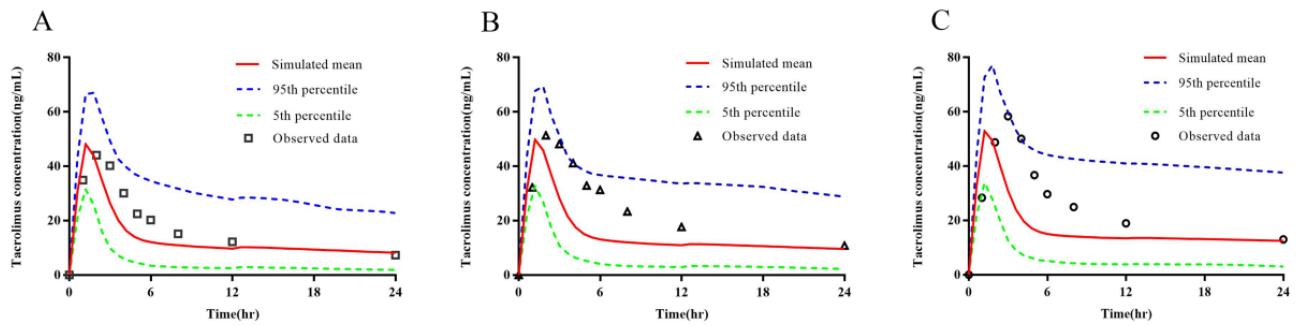

FIGURE 3 - Simulation of tacrolimus whole-blood concentration-time profiles after a 3mg single oral dose in combination with 200mg voriconazole twice daily at steady state in CYP2C19 EMs(A), IMs(B), and PMs(C). Red line represents the mean value of the simulated population. Blue and green line represent the 5th-95th percentiles value of simulated population. Squires, triangles, and circles represent the EMs, IMs and PMs observed data respectively.

TABLE IV - The predicted versus observed pharmacokinetic parameters of tacrolimus coadministered with voriconazole according to CYP2C19 genotypes

CYP2C19 Genotype	AUC0-24(ng·h/mL)			Cmax(ng/mL)			Tmax(h)		
	Predicted	Observed	Fold error	Predicted	Observed	Fold error	Predicted	Observed	Fold error
EM	328.03	389.5	1.19	49.38	48.3	1.02	1.39	2.3	1.65
IM	357.70	540.6	1.51	51.29	54.9	1.07	1.43	2.3	1.61
PM	415.4	570.5	1.37	54.74	60.5	1.11	1.46	2.8	1.92

Simulating the disappearance of the DDIs after voriconazole withdrawal

Simulating the clinical scenes according to the study design, tacrolimus whole blood concentration increased immediately after taking voriconazole in all three populations. With the discontinuation of voriconazole, it took 146h(6.08d), 90h(3.75d) and 66h(2.75d) to make the tacrolimus in vivo exposure return to the baseline in PMs, IMs and EMs, respectively (Figure 4).

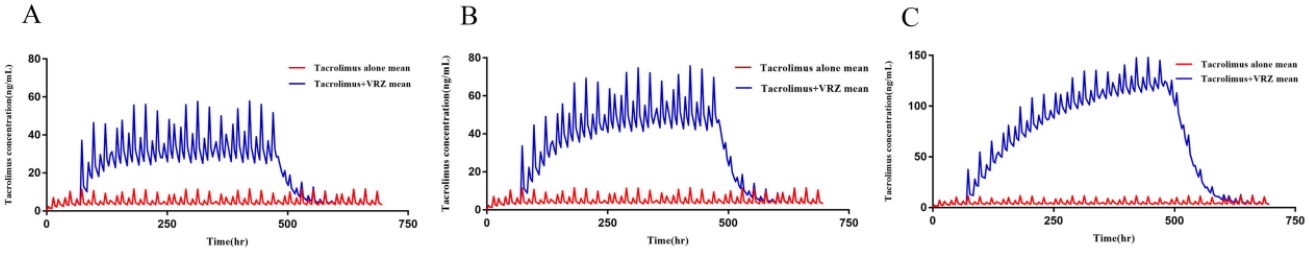

FIGURE 4 - Simulation of the disappearance of the DDIs after voriconazole withdrawal in CYP2C19 EMs(A), IMs(B) and PMs(C). Red line represents the mean value of tacrolimus concentration when tacrolimus used alone. Blue line represents the mean value of tacrolimus concentration when tacrolimus used with and without voriconazole.

DISSCUSSION

In solid organ transplant (SOT) recipients, the likelihood of invasive aspergillus infection occurs from 0.1% to 3.5%, depending on the specific type of transplant and ethic. Invasive aspergillus infection accounts for 18-30% of invasive fungal disease(IFD) of all SOT recipients. It also has a high mortality, especially in the patients who develop invasive pulmonary diseases, with the mortality up to 67-82% (Gavalda *et al*., 2014). Voriconazole is recommended as the primary treatment of invasive aspergillosis (IA) (Patterson *et al*., 2016). As a result, coadministration of tacrolimus and voriconazole is common in clinical practice. The drug interaction between voriconazole and tacrolimus has clinical significance, and the manufacturer recommends reducing the daily tacrolimus dose to one-third when coadministered with voriconazole. According to our clinical practice experience and some case reports, this reduction of tacrolimus may not be sufficient. Drik R Kupers et al. (Kuypers *et al*., 2006) reported a renal allograft patient with Aspergillus fumigatus infection, who was treated by voriconazole, had to taper the tacrolimus dose from 1.5mg daily to 0.5mg every third day to maintain the trough concentration between 4-6ng/mL. Another case report showed the coadministration of voriconazole and tacrolimus led to a 10-fold increase in tacrolimus trough concentration in a liver transplant recipient(Venkataramanan, 2002). Likewise, Eisuke Mochizuki et al. (Mochizuki *et al*., 2015) reported that a dermatomyositis-associated interstitial lung disease patient receiving tacrolimus eventually reduced 85% maintenance dose after initiating voriconazole. Even in one case reported by D. Capone MD et al. (Capone *et al*., 2010), a kidney transplant patient had to discontinue the immunosuppressant drug. In general, pharmacokinetic interactions commonly occur via metabolism enzymes or drug transporters. Tacrolimus is extensively metabolized by *CYP3A4* and transported by P-glycoprotein (P-gp). Voriconazole is neither substrate nor inhibitor of P-gp (Saad, DePestel, Carver, 2006), and it only inhibits *CYP3A4*. Many types of research have revealed that voriconazole increases the serum concentration of tacrolimus via competitive inhibition of CYP3A4 (Trifilio *et al*., 2010). Meanwhile, voriconazole serum concentration is associated with the *CYP2C19* genotype. Based on the information mentioned above, it could be hypothesized that the *CYP2C19* polymorphism, which determines the exposure and concentration of voriconazole, might affect the magnitude of DDIs between tacrolimus and voriconazole. It might explain why the fixed-dose reduction cannot be satisfactory as well.

Among over thirty alleles, the *CYP2C19* gene exhibits significant differences among racial groups. The two most common nonfunctional alleles are *CYP2C19*2* and *CYP2C19*3*, which account for >99% of Oriental PM alleles (Ferguson *et al*., 1998). Approximately 12-23% of Asians, 7% of African Americans, 3-5% of Caucasians, and 0.9% of Hispanics are *CYP2C19* PMs (Moriyama *et al*., 2015), indicating that they will achieve higher voriconazole exposure than EMs and IMs do. Due to the high ratio of *CYP2C19* PMs in Asians, the development of PBPK models based on *CYP2C19* genotypes is especially meaningful to Asian patients. A relatively novel *CYP2C19* gene allele, *CYP2C19*17*, associated with increased *CYP2C19* expression and catalytic activity has been identified (Sim *et al*., 2006). The individuals who express *CYP2C19*17* allele are defined as ultra-rapid metabolizers (URMs). However, *CYP2C19*17* occurs infrequently in Asians with 0.15 to 0.44% prevalence (Hirota, Eguchi, Ieiri, 2013). Hence this study did not consider the URMs. Based on the initial model, we used the "top-down" model building method to optimize the metabolic parameters, such as V_{max} and enzyme abundance. The fold error between the predicted and observed value was less than 2, indicating that the modified PBPK model well-described regarding the physiological disposition of voriconazole in different metabolizing populations and can be applied in the prediction of DDIs. By our PBPK models, the predicted voriconazole AUC_{0-12}/C_{max} of the PMs and IMs was 3.51/2.66 and 1.54/1.37 times higher than EMs, respectively. This agreed with several clinical studies' findings, which reported the AUC of PMs was about 3 times higher than Ems (Lee *et al*., 2012; Scholz *et al*., 2009). Infectious Diseases Society of America (IDSA) guideline recommends on voriconazole trough level > 1-1.5 μg/mL for efficacy and < 5-6μg/mL to minimize toxicity (Patterson *et al*., 2016). Though

regular pharmacogenomic screening is not recommended, clinicians should consider the impact of *CYP2C19* polymorphism if the desired therapeutic effect is not achieved or unexpected toxicity occurred with standard regimen without any other risk factors (pathogens, drug resistance, etc.).

With a narrow therapeutic index, tacrolimus exhibits a tremendous pharmacokinetic variability among both interindividual and intraindividual subjects (Venkataramanan *et al.*, 1995). The simulated pharmacokinetic profiles were comparable to the observed data, which indicated that the optimized model could reasonably predict the pharmacokinetics of tacrolimus and the associated DDIs. Furthermore, besides the substrate, namely tacrolimus, the optimized model can load three different inhibitors simultaneously when predicting DDIs at one time. In fact, most SOT recipients take more than one drug every day. Thus, the optimized model also provides a tool for more detailed research on DDIs.

After the performance of the refined PBPK models was approved, the simulation of DDIs was carried out. FE value of AUC_{0-24}, C_{max} and T_{max} were all within 2 folds. As AUC_{0-24} is generally used to explain the physiological drug disposition, $R_{predicted/observed}$ of AUCR was introduced to further improve the performance of the optimized PBPK models, and the $R_{predicted/observed}$ of AUCR were all between 0.5-2. The results indicated that the models could well describe the DDIs between tacrolimus and voriconazole of different *CYP2C19* genotype. The prescribing information of tacrolimus (Prograf®) mentions "repeating oral dose administration of voriconazole increased tacrolimus (0.1mg/kg single dose) AUC_{τ} in healthy subjects by an average of 3-fold." However, according to our simulation, when coadministering tacrolimus with voriconazole, the AUC_{0-24} of tacrolimus was increased 3.03, 3.31 and 3.84 times in EMs, IMs and PMs, respectively. It may explain that the current "one size fits all" dose reduction was not applicable to all patients and these patients need more than 2/3 dose reduction in most cases. Hence it is necessary to consider the impact of *CYP2C19* polymorphism and modify the dose according to the *CYP2C19* gene status, when initiating voriconazole therapy in patients taking tacrolimus. Dose-dependent autoinhibition is proposed as underlying mechanism for voriconazole's nonlinear pharmacokinetics. Thus, the clinicians should take the infusion rate of vorconazole injection into consideration in clinical practice. It was recommended that the infusion rate of voriconazole injection should be 2 h (Hohmann *et al.*, 2017).

Most SOT recipients should receive immunosuppression therapy throughout their life for the reason of allograft rejection (Khwaja, 2010; Shi, 2016). While the antifungal infection is relatively temporary, when voriconazole is discontinued, the tacrolimus dose should be increased as necessary. Excessive dose modification of tacrolimus may lead to high blood concentration and increase the occurrence of the serious adverse reaction, such as nephrotoxicity, hypertension, and hyperglycemia. On the contrary, the risk of organ rejection at low blood concentration may be higher due to an untimely dose increase. A previous research (Kramer *et al.*, 2011) documented an acute rejection occurred in a lung transplant recipient due to rapid drop of tacrolimus level after itraconazole withdrawal. According to our literature research, little information is available regarding the duration of DDI between voriconazole and tacrolimus when voriconazole was discontinued and how the tacrolimus dose should be adjusted accordingly. It was mentioned that at least 7-10 days were required for concentrations of immunosuppressant to return to the baseline after an azole withdrawal (Saad, DePestel, Carver, 2006). D. Capone MD et al. (Capone *et al.*, 2010) noted the clinical phenomenon that the time of appearance of DDI (1 day) was faster than that necessary for its disappearance (8 days after voriconazole discontinuation). Both overexposure and underexposure of tacrolimus can put SOT recipients at risks. The results of our PBPK models simulation demonstrated the appearance of DDI occurs immediately after voriconazole taking in all three different genotypes, which indicated instant dose reduction was required. Duration of tacrolimus in vivo exposure after voriconazole discontinuation varied in different populations (6.08d for PMs, 3.75d for IMs and 2.75d for EMs) due to *CYP2C19* polymorphism. According to our simulation, in order to keep the trough blood concentration of tacrolimus within the target range, the clinicians should increase tacrolimus dose to the initial

level no more than 6 days after voriconazole removed from the regimen, and as to IMs and EMs the time could be sooner. Meanwhile, intensive monitoring of tacrolimus whole blood concentration should be carried out as well, especially in the early stages of drug treatment changes. The dose of tacrolimus is generally determined by the drug target level, which varies with the type of transplant organ and the time after transplant (Khwaja, 2010; Lucey *et al.*, 2013; Shi, 2016), so clinicians should verify the exact tacrolimus target whole blood concentration when discontinuing voriconazole for each patient. Tacrolimus dosage can be calculated according to the target whole blood concentration by the PBPK modles. Especially in those whose tacrolimus whole blood drug centration cannot reach the target range after frequently empirical dose adjustments. Furthermore, a lot of data about the relationship between tacrolimus blood concentrations and its efficacy or toxicity has been collected in the clinical practice. The combination of these reported exposure-response correlations with PBPK modeling of tarolimus DDI may utilize the strategy which has been employed to investigate the drug exposure and hepatotoxicity (Albrecht *et al.*, 2019; Li *et al.*, 2021).

CONCLUSION

In conclusion, there is a clinically significant increase of tacrolimus blood concentration when coadministering with voriconazole, and *CYP2C19* genotype is one of the determining factors of the magnitude of DDIs even though tacrolimus is mainly metabolized by *CYP3A*. For the reason of much higher PM population than other ethnic groups, considering *CYP2C19* polymorphism when adjusting tacrolimus dose both initiating and discontinuing voriconazole has more clinical values in Asians. The developed and optimized PBPK models can represent tools to be applied to assist the precise dosing of tacrolimus for SOT patients with secondary IA and after the end of IA treatment.

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REFERENCES

Albrecht W, Kappenberg F, Brecklinghaus T, Stoeber R, Marchan R, Zhang M, et al. Prediction of human drug-induced liver injury (DILI) in relation to oral doses and blood concentrations. Arch Toxicol. 2019;93(6):1609-1637.

Capone D, Tarantino G, Gentile A, Sabbatini M, Polichetti G, Santangelo M, et al. Effects of voriconazole on tacrolimus metabolism in a kidney transplant recipient. J Clin Pharm Ther. 2010;35(1):121-124.

Chan TS, Yu H, Moore A, Khetani SR, Tweedie D. Meeting the challenge of predicting hepatic clearance of compounds slowly metabolized by cytochrome P450 using a novel hepatocyte model, HepatoPac. Drug Metab Dispos. 2013;41(12):2024-2032.

Damle B, Varma MV, Wood N. Pharmacokinetics of voriconazole administered concomitantly with fluconazole and population-based simulation for sequential use. Antimicrob Agents Chemother. 2011;55(11):5172-5177.

Dresser GK, Spence JD, Bailey DG. Pharmacokinetic-pharmacodynamic consequences and clinical relevance of cytochrome P450 3A4 inhibition. Clin Pharmacokinet. 2000;38(1):41-57.

Drug interaction studies- study design da, and clinical implications guidence for industry (draft gudience). Access October 10, 2019.

Ferguson RJ, De Morais SM, Benhamou S, Bouchardy C, Blaisdell J, Ibeanu G, et al. A new genetic defect in human CYP2C19: mutation of the initiation codon is responsible for poor metabolism of S-mephenytoin. J Pharmacol Exp Ther. 1998;284(1):356-361.

Gavalda J, Meije Y, Fortun J, Roilides E, Saliba F, Lortholary O, et al. Invasive fungal infections in solid organ transplant recipients. Clin Microbiol Infect. 2014;20(Suppl 7):27-48.

Grimstein M, Yang Y, Zhang X, Grillo J, Huang SM, Zineh I, et al. Physiologically based pharmacokinetic modeling in regulatory science: An update from the U.S. food and drug administration's office of clinical pharmacology. J Pharm Sci. 2019;108(1):21-25.

Guideline on the Investigation of Drug Interact ions. Access October 10, 2019.

Hirota T, Eguchi S, Ieiri I. Impact of genetic polymorphisms in CYP2C9 and CYP2C19 on the pharmacokinetics of clinically used drugs. Drug Metab Pharmacokinet. 2013;28(1):28-37.

Hohmann N, Kreuter R, Blank A, Weiss J, Burhenne J, Haefeli WE, et al. Autoinhibitory properties of the parent but not of the N-oxide metabolite contribute to infusion rate-dependent voriconazole pharmacokinetics. Br J Clin Pharmacol. 2017;83(9):1954-1965.

https://www.pharmvar.org/htdocs/archive/cyp2c19.htm. Access March 30, 2021.

Husain S, Camargo JF. Invasive Aspergillosis in solid-organ transplant recipients: Guidelines from the american society of transplantation infectious diseases community of practice. Clin Transplant. 2019;33(9):e13544.

Hyland R, Jones BC, Smith DA. Identification of the cytochrome P450 enzymes involved in the N-oxidation of voriconazole. Drug Metab Dispos. 2003;31(5):540-547.

Imamura CK, Furihata K, Okamoto S, Tanigawara Y. Impact of cytochrome P450 2C19 polymorphisms on the pharmacokinetics of tacrolimus when coadministered with voriconazole. J Clin Pharmacol. 2016;56(4):408-413.

Jeong S, Nguyen PD, Desta Z. Comprehensive in vitro analysis of voriconazole inhibition of eight cytochrome P450 (CYP) enzymes: major effect on CYPs 2B6, 2C9, 2C19, and 3A. Antimicrob Agents Chemother. 2009;53(2):541-551.

Khwaja A. KDIGO guidelines for care of the kidney transplant recipient. Nephron Clin Pract. 2010;116(1):c27-28.

Kramer MR, Amital A, Fuks L, Shitrit D. Voriconazole and itraconazole in lung transplant recipients receiving tacrolimus (FK 506): efficacy and drug interaction. Clin Transplant. 2011;25(2):E163-167.

Kuypers DR, Claes K, Evenepoel P, Vanrenterghem Y. Clinically relevant drug interaction between voriconazole and tacrolimus in a primary renal allograft recipient. Transplantation. 2006;81(12):1750-1752.

Lee S, Kim BH, Nam WS, Yoon SH, Cho JY, Shin SG, et al. Effect of CYP2C19 polymorphism on the pharmacokinetics of voriconazole after single and multiple doses in healthy volunteers. J Clin Pharmacol. 2012;52(2):195-203.

Li S, Yu Y, Bian X, Yao L, Li M, Lou YR, et al. Prediction of oral hepatotoxic dose of natural products derived from traditional Chinese medicines based on SVM classifier and PBPK modeling. Arch Toxicol. 2021;95(5):1683-1701.

Lucey MR, Terrault N, Ojo L, Hay JE, Neuberger J, Blumberg E, et al. Long-term management of the successful adult liver transplant: 2012 practice guideline by the American Association for the Study of Liver Diseases and the American Society of Transplantation. Liver Transpl. 2013;19(1):3-26.

Mochizuki E, Furuhashi K, Fujisawa T, Enomoto N, Inui N, Nakamura Y, et al. A case of treatment with voriconazole for chronic progressive pulmonary aspergillosis in a patient receiving tacrolimus for dermatomyositis-associated interstitial lung disease. Respir Med Case Rep. 2015;16:163-165.

Mori T, Kato J, Yamane A, Sakurai M, Kohashi S, Kikuchi T, et al. Drug interaction between voriconazole and tacrolimus and its association with the bioavailability of oral voriconazole in recipients of allogeneic hematopoietic stem cell transplantation. Int J Hematol. 2012;95(5):564-569.

Moriyama B, Kadri S, Henning SA, Danner RL, Walsh TJ, Penzak SR. Therapeutic drug monitoring and genotypic screening in the clinical use of voriconazole. Curr Fungal Infect Rep. 2015;9(2):74-87.

Patterson TF, Thompson GR, 3rd, Denning DW, Fishman JA, Hadley S, Herbrecht R, et al. Practice Guidelines for the diagnosis and management of Aspergillosis: 2016 Update by the infectious diseases society of america. Clin Infect Dis. 2016;63(4):e1-e60.

Pfizer. Vfend Prescribing Information in U.S.: voriconazole tablets, injection suspensions. 2014.

Poulin P, Theil FP. Prediction of pharmacokinetics prior to in vivo studies. II. Generic physiologically based pharmacokinetic models of drug disposition. J Pharm Sci. 2002;91(5):1358-1370.

Purkins L, Wood N, Ghahramani P, Greenhalgh K, Allen MJ, Kleinermans D. Pharmacokinetics and safety of voriconazole following intravenous- to oral-dose escalation regimens. Antimicrob Agents Chemother. 2002;46(8):2546-2553.

Saad AH, DePestel DD, Carver PL. Factors influencing the magnitude and clinical significance of drug interactions between azole antifungals and select immunosuppressants. Pharmacotherapy. 2006;26(12):1730-1744.

Scholz I, Oberwittler H, Riedel KD, Burhenne J, Weiss J, Haefeli WE, et al. Pharmacokinetics, metabolism and bioavailability of the triazole antifungal agent voriconazole in relation to CYP2C19 genotype. Br J Clin Pharmacol. 2009;68(6):906-915.

Shi B Y ea. Chinese guideline for diagnosis, prevention, and treatmen of invasive fungal infections in solid organ transplant recipients. Chin J Organ Transplant. 2016;37(5):300-305.

Sim SC, Risinger C, Dahl ML, Aklillu E, Christensen M, Bertilsson L, et al. A common novel CYP2C19 gene variant causes ultrarapid drug metabolism relevant for the drug response to proton pump inhibitors and antidepressants. Clin Pharmacol Ther. 2006;79(1):103-113.

Staatz CE, Tett SE. Clinical pharmacokinetics and pharmacodynamics of tacrolimus in solid organ transplantation. Clin Pharmacokinet. 2004;43(10):623-653.

Trifilio SM, Scheetz MH, Pi J, Mehta J. Tacrolimus use in adult allogeneic stem cell transplant recipients receiving voriconazole: preemptive dose modification and therapeutic drug monitoring. Bone Marrow Transplant. 2010;45(8):1352-1356.

Venkataramanan R, Swaminathan A, Prasad T, Jain A, Zuckerman S, Warty V, et al. Clinical pharmacokinetics of tacrolimus. Clin Pharmacokinet. 1995;29(6):404-430.

Venkataramanan R, Zang S, Gayowski T, Singh N. Voriconazole inhibition of the metabolism of tacrolimus in a liver transplant recipient and in human liver microsomes. Antimicrob Agents Chemother. 2002;46(9):3091-3093.

Vieira MD, Kim MJ, Apparaju S, Sinha V, Zineh I, Huang SM, et al. PBPK model describes the effects of comedication and genetic polymorphism on systemic exposure of drugs that undergo multiple clearance pathways. Clin Pharmacol Ther. 2014;95(5):550-557.

Zhang H, Bu F, Li L, Jiao Z, Ma G, Cai W, et al. Prediction of drug-drug interaction between tacrolimus and principal ingredients of wuzhi capsule in chinese healthy volunteers using physiologically-based pharmacokinetic modelling. Basic Clin Pharmacol Toxicol. 2018;122(3):331-340.

Zhang S, Pillai VC, Mada SR, Strom S, Venkataramanan R. Effect of voriconazole and other azole antifungal agents on CYP3A activity and metabolism of tacrolimus in human liver microsomes. Xenobiotica. 2012;42(5):409-416.

Zhou Y, Ingelman-Sundberg M, Lauschke VM. Worldwide distribution of cytochrome p450 alleles: a meta-analysis of population-scale sequencing projects. Clin Pharmacol Ther. 2017;102(4):688-700.

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