Original Article

Diltiazem augments the influence of MDR1 genotype status on cyclosporine concentration in Chinese patients with renal transplantation

Yi-xi WANG^{1,2, #}, Jia-li LI^{1, #}, Xue-ding WANG¹, Yu ZHANG¹, Chang-xi WANG^{3, *}, Min HUANG^{1, *}

¹Institute of Clinical Pharmacology, School of Pharmaceutical Sciences, Sun Yat-sen University, Guangzhou 510080, China;

²Department of Pharmacy, The Second Affiliated Hospital of Guangzhou Medical University, Guangzhou 510260, China;

³Department of Organ Transplant, The First Affiliated Hospital, Sun Yat-sen University, Guangzhou 510080, China

Aim: Co-administration of diltiazem can reduce the dosage of cyclosporine (CsA) in patients with renal transplantation. In this study, we investigated how diltiazem altered the relationship between MDR1 genetic polymorphisms and CsA concentration in Chinese patients with renal transplantation.

Methods: A total of 126 renal transplant patients were enrolled. All the patients received CsA (2–4 mg/kg⁻¹·d⁻¹), and diltiazem (90 mg/d) was co-administered to 76 patients. MDR1-C1236T, G2677T/A, and C3435T polymorphisms were genotyped. The whole blood concentration was measured using the FPIA method, and the adjusted trough concentrations were compared among the groups with different genotypes.

Results: In all patients, MDR1-C1236T did not influence the adjusted CsA trough concentration. With regard to MDR1-3435, the adjusted CsA trough concentration was significantly higher in TT carriers than in CC and CT carriers when diltiazem was co-administered (58.83±13.95 versus 46.14±7.55 and 45.18±12.35 ng/mL per mg/kg, $P=0.011$), and the differences were not observed in patients without diltiazem co-administered. With regard to MDR1-2677, the adjusted CsA trough concentration was significantly higher in TT carriers than in GG and GT carriers when diltiazem was co-administered (61.31±12.93 versus 52.25±7.83 and 39.70±7.26 ng/mL per mg/kg, $P=0.0001$). The differences were also observed in patients without diltiazem co-administered (43.27±5.95 versus 35.22±7.55 and 29.54±5.35 ng/mL per mg/kg, $P=0.001$). The adjusted CsA trough blood concentration was significantly higher in haplotype T-T-T and haplotype T-T-C carriers than in non-carriers, regardless of diltiazem co-administered.

Conclusion: MDR1 variants influence the adjusted CsA trough concentration in Chinese patients with renal transplant, and the influence more prominent when diltiazem is co-administered.

Keywords: cyclosporin; diltiazem; MDR1; polymorphisms; genotype; renal transplantation; Chinese patients

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Introduction

Cyclosporine (CsA), a calcineurin inhibitor, is widely used to prevent acute rejection after solid organ transplantation^[1]. However, cyclosporine has low oral bioavailability, a narrow therapeutic index and shows marked interindividual differences in pharmacokinetics^[2]. The ATP-driven efflux pump, P-glycoprotein (P-gp), has been identified as an absorptive barrier to orally administered CsA. Therefore, P-gp might play a role in the disposition of CsA^[3].

The multidrug resistance gene (MDR1), encoding P-gp, is expressed at high levels in the adrenal glands and kidneys, at intermediate levels in the lung, liver, lower jejunum, colon and rectum, and at low levels in many other tissues^[4]. As a transporter, P-gp plays a significant role in drug disposition, *ie*, absorption, distribution, and excretion, and might also be involved in the secretion of steroids^[5]. A total of 50 single nucleotide polymorphisms (SNPs) have been identified in MDR1, including C1236T, G2677T/A and C3435T in exons 12, 21 and 26, respectively, and these functionally important mutations can form different haplotypes. Both SNPs and haplotypes have been demonstrated as highly polymorphic among individuals and different ethnic groups^[6–8]. The genetic polymorphisms of MDR1 have been implicated as one of the factors resulting in CsA pharmacokinetic variation.

[#] These authors contributed equally to this work.

^{*} To whom correspondence should be addressed.

E-mail huangmin@mail.sysu.edu.cn (Min HUANG);

wcx6363@163.com (Chang-xi WANG)

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Co-administration with diltiazem has been frequently used, and this treatment might have beneficial effects beyond the economic impact associated with the dose reduction of CsA. Indeed, diltiazem is relatively safe, showing a useful anti-hypertensive action, potentially exhibiting blood pressure control and renal protection^[9]. It has been reported that the CsA dosage was 12% lower in the diltiazem group than that in the non-diltiazem group at one year after transplantation. Furthermore, the diltiazem group might be associated with significantly lower probability to develop chronic allograft nephropathy than the non-diltiazem group^[10].

However, the correlation between MDR1 genetic polymorphisms and cyclosporine concentration when diltiazem is co-administered remains unclear. Therefore, in the present study, we retrospectively compared the impact of the MDR1 SNP/haplotype on cyclosporine concentration with and without diltiazem co-administration to assess the influence of diltiazem on the association of MDR1 genetic polymorphisms with cyclosporine concentration.

Materials and methods

Patients

This study was conducted between June 2008 and December 2011, involving a total of 126 renal transplant recipients (82 males, 44 females) who underwent transplantation at the Department of Organ Transplant, The First Affiliated Hospital, Sun Yat-sen University and were enrolled during outpatient visits at the Renal Transplant Clinic, The First Affiliated Hospital, Sun Yat-sen University. The average age of the patients was 29.15±15.36 years (range, 18–74 years), and the average body weight was 56.65±9.82 kg. All patients were maintained on a triple immunosuppressive regimen comprising CsA, mycophenolate mofetil and steroids, and the average time of post-transplantation was 25 months. During the experimental period, oral prednisolone was administered at 10 mg/d and mycophenolate mofetil was administered at 1 g bid. Diltiazem, as a CsA-sparing agent, was administered at 90 mg/d as a single daily dose to 76 patients. The patients did not receive any other drugs, such as calcium channel blockers (nicardipine and verapamil), antiepileptics (phenytoin and carbamazepine), antimycotics (fluconazole and ketoconazole), or macrolide antibiotics (erythromycin and clarithromycin), which interact with CsA. Patients fulfilling the above criteria were included. This study was performed in accordance with the Declaration of Helsinki, and ethical approval was obtained from the Ethical Committee of Sun Yat-sen University, Guangzhou, China. Written informed consent was obtained from all subjects.

CsA dosage and quantitation

The dosage of CsA was 2–4 mg·kg⁻¹·d⁻¹ and the daily dosage was adjusted, according to the blood trough CsA concentration (C₀), to a target concentration of 100–120 ng/mL. The body weight, CsA dosage, and whole blood concentration were recorded at 5 d after the patient was administered the same dosage of CsA.

CsA was administered daily, in equal amounts, at 8:00 AM

and 8:00 PM. To determine the trough concentration (C₀), blood samples (using ethylenediaminetetraacetic acid as an anticoagulant) were collected at 8:00 AM, prior to administering the morning dose. The samples were assayed using the commercially available CsA whole blood monoclonal antibody fluorescence polarization assay (FPIA; TDx; Abbott Laboratories, Chicago, IL, USA)^[11]. The weight-adjusted CsA dosage (mg·kg⁻¹·d⁻¹) and the adjusted concentration (ng/mL per mg·kg⁻¹·d⁻¹) were calculated.

Genotyping the MDR1 polymorphism

Total DNA was extracted from the peripheral leukocytes obtained from the subjects using the phenol-chloroform extraction method as previously described^[12]. Polymerase chain reaction, followed by restriction fragment length polymorphism analysis (PCR-RFLP) was used to genotype the MDR1 polymorphisms, with only slight modifications^[13, 14]. Details regarding the primer sequences and restriction enzymes used in the present study are shown in Table 1.

Table 1. Primers and restriction enzyme used to detect the mutations in MDR1 gene.

Primers	5'-3' Sequence	Exon	Enzyme
MDR1-1236-F	TACCCATCTCGAAAAGAAGTTAAGG	12	
MDR1-1236-R	GAAAGATGTGAATGTGACTGCTGAT	12	<i>Hae</i> I
MDR1-2677-F	TGCAGGCTATAGTTCCAGG	21	
MDR1-2677-AR	GTTTGACTCACCTTCCCAG	21	<i>Bsr</i> I
MDR1-2677-TR	TTAGTTTGACTCACCTTCCCG	21	<i>Ban</i> I
MDR1-3435-F	TGCTGGTCCTGAAGTTGATCTGTGAAC	26	
MDR1-3435-R	ACATTAGGCAGTGACTCGATGAAGGCA	26	<i>Mbo</i> III

Statistical analysis

The data were analyzed using the computer software SPSS (Statistical Package for the Social Sciences) for Windows (Version 12.0, Chicago, IL, USA). The MDR1 1236-2677-3435 haplotype analysis was performed using PHASE 2.1 software (downloaded from <http://www.stat.washington.edu/stephens/phase/download.html>). The adjusted trough blood concentration (ng/mL per mg/kg) and daily dose (mg/kg) required to achieve target blood concentrations were compared among individuals according to the allelic status of MDR1. The quantitative variables are expressed as the mean±standard deviation (SD). The distribution of quantitative parameters was compared between groups using parametric or nonparametric tests depending on the normality of the variables tested (Wilks-Shapiro test). For each analysis, *P* values less than 0.05 were considered statistically significant.

Results

Influence of diltiazem on CsA trough and adjusted trough concentrations

One hundred twenty-six Chinese renal transplant patients

were enrolled in this study and divided into Dil(+) ($n=76$) and Dil(-) ($n=50$) groups (Table 2). As shown in Table 3, significantly different daily dosages were observed between Dil(+) and Dil(-) groups (2.76 ± 0.78 versus 3.61 ± 0.97 mg/kg, $P<0.001$). The adjusted CsA trough concentrations were significantly

Table 2. Patients background data.

Total: 126	With diltiazem (Dil+)	Without diltiazem (Dil-)
<i>n</i>	76	50
Age		
Range (years)	21-70	18-74
Median (years)	41.5	39.5
Sex		
Male	51	31
Female	25	19
Time post-transplant		
Range (months)	30.1 $\pm$ 13.2	28.9 $\pm$ 14.6
Median (months)	25	24.5
Body weight		
Weight range (kg)	42-80	40-84
Weight median (kg)	58.8	55
Drug administration		
Cyclosporin	2-4 mgkg ⁻¹ d ⁻¹	
Mycophenolate mofetil (g/d)	2	2
Prednisone (mg/d)	10	10
Diltiazem (mg/d)	90	NO

Table 3. Difference of CsA dose, CsA concentration and adjusted CsA blood concentration between Dil(+) patients and Dil(-) patients.

<i>n</i> =126	Dil(+)	Dil(-)
	<i>n</i> =76	<i>n</i> =50
CsA daily dose (mgkg ⁻¹ d ⁻¹)	2.76 $\pm$ 0.78	3.61 $\pm$ 0.97
	$P=0.000$	
CsA trough concentration (ng/mL)	125.99 $\pm$ 29.61	114.38 $\pm$ 23.60
	$P=0.022$	
Dose adjusted concentration (ng/mL per mg/kg)	47.32 $\pm$ 11.71	32.87 $\pm$ 7.62
	$P=0.000$	

higher in the Dil(+) group than in the Dil(-) group during the stable stage in Chinese renal transplant patients (47.32 ± 11.71 versus 32.87 ± 7.62 ng/mL per mg/kg, $P<0.001$).

Influence of MDR1 genotypes on CsA trough and adjusted trough concentrations

The distribution of the MDR1 C1236T, G2677T/A, and C3435T alleles was consistent with Hardy-Weinberg equilibrium (each $P>0.05$). As shown in Table 4, there were no significant differences in the CsA trough concentrations, adjusted trough concentrations and daily dosage among different MDR1 C1236T genotype groups in both patients who used diltiazem and those who did not use diltiazem during the stable stage.

As shown in Figure 1, 2, and Table 5, the adjusted CsA trough concentration was significantly higher in MDR1-2677TT carriers than in GG and GT carriers in the Dil(+) group, showing 61.31 ± 12.93 versus 52.25 ± 7.83 and 39.70 ± 7.26 ng/mL per mg/kg, respectively ($P=0.004$). Moreover, these

Table 4. Difference of CsA dose, CsA concentration and adjusted CsA blood concentration among MDR1-1236 genotype in Dil(+) patients or Dil(-) patients.

Dil(+) (<i>n</i> =76)	CC	MDR1-1236 CT	TT
Patients number, <i>n</i>	10	31	35
CsA daily dose (mgkg ⁻¹ d ⁻¹)	2.72 $\pm$ 0.54	2.73 $\pm$ 0.60	2.81 $\pm$ 0.98
		$P=0.987$	
CsA trough concentration (ng/mL)	126.58 $\pm$ 29.07	129.73 $\pm$ 29.22	122.06 $\pm$ 30.53
		$P=0.350$	
Dose adjusted concentration (ng/mL per mg/kg)	46.80 $\pm$ 7.46	48.39 $\pm$ 10.04	46.21 $\pm$ 13.46
		$P=0.826$	
Dil (-) (<i>n</i> =50)	CC	MDR1-1236 CT	TT
Patients number, <i>n</i>	9	14	27
CsA daily dose (mgkg ⁻¹ d ⁻¹)	3.82 $\pm$ 0.92	3.98 $\pm$ 0.59	3.35 $\pm$ 0.75
		$P=0.163$	
CsA trough concentration (ng/mL)	118.75 $\pm$ 33.04	119.29 $\pm$ 19.64	110.78 $\pm$ 21.27
		$P=0.442$	
Dose adjusted concentration (ng/mL per mg/kg)	31.50 $\pm$ 7.59	31.50 $\pm$ 8.79	33.83 $\pm$ 7.79
		$P=0.531$

Figure 1. Correlation of MDR1-2677 genotype with the dose-adjusted trough concentration of cyclosporine in patients who co-administered with diltiazem. Dose-adjusted trough concentration of cyclosporine was significantly higher in MDR1-2677TT carriers than that in GG plus GT carriers. *A=GA+AA+AT.

Figure 2. Correlation of MDR1-2677 genotype with the dose-adjusted trough concentration of cyclosporine in patients who didn't use diltiazem. Dose-adjusted trough concentration of cyclosporine was significantly higher in MDR1-2677TT carriers than that in GG plus GT carriers. *A=GA+AA+AT.

differences were also observed in the Dil(-) group, showing 43.27 ± 5.95 versus 35.22 ± 7.55 plus 29.54 ± 5.35 ng/mL per mg/kg, respectively ($P=0.001$). The adjusted CsA trough concentrations were increased with the increasing number of 2677-T alleles in all patients during the stable stage after renal transplantation.

As shown in Figure 3 and Table 6, the adjusted CsA trough concentrations were significantly higher in MDR1-3435TT carriers than in CC and CT carriers in the Dil(+) group, showing 58.83 ± 13.95 versus 46.14 ± 7.55 and 45.18 ± 12.35 ng/mL per mg/kg, respectively ($P=0.011$). However, no significant difference was observed in the CsA trough concentrations, adjusted trough concentrations and daily dosage among MDR1 C3435T genotype groups in patients who did not use diltiazem.

Influence of MDR1 haplotype on CsA adjusted trough concentrations

The most common MDR1 1236-3677-3435 haplotype was T-G-C, with a frequency of 31.2%. Other haplotypes were also detected, including C-G-C, T-T-T, T-G-T, and T-T-C, with frequencies of 26.2%, 18.7%, 10.3%, and 5.2%, respectively.

As shown in Table 7, in the Dil(+) group, significantly higher adjusted CsA trough concentrations were observed in carriers of haplotypes T-T-T and T-T-C compared with non-carriers ($P=0.007$ and 0.001 , respectively), while haplotype T-G-C carriers had significantly lower adjusted CsA trough concentrations than non-carriers ($P=0.0001$). However, in the Dil(-)

Table 5. Difference of CsA dose, CsA concentration and adjusted CsA blood concentration among MDR1-2677 genotype in Dil(+) patients or Dil(-) patients. *A=GA+AA+AT.

Dil(+) (n=76)	MDR1-2677			
	GG	GT	TT	*A
Patients number, n	38	24	11	3
CsA daily dose ($\text{mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$)	2.96 ± 0.92	2.75 ± 0.52	2.35 ± 0.52	1.93 ± 0.19
$P=0.017$				
CsA trough concentration (ng/mL)	113.38 ± 25.75	142.16 ± 26.89	140.81 ± 27.37	101.87 ± 17.40
$P=0.001$				
Dose adjusted concentration (ng/mL per mg/kg)	39.70 ± 7.26	52.25 ± 7.83	61.31 ± 12.93	53.15 ± 10.37
$P=0.0001$				
Dil(-) (n=50)	MDR1-2677			
	GG	GT	TT	*A
Patients number, n	27	15	6	2
CsA daily dose ($\text{mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$)	3.18 ± 0.68	3.95 ± 0.66	2.87 ± 0.73	4.43
$P=0.004$				
CsA trough concentration (ng/mL)	115.35 ± 26.15	109.57 ± 21.02	123.99 ± 16.23	109.35
$P=0.569$				
Dose adjusted concentration (ng/mL per mg/kg)	29.54 ± 5.35	35.22 ± 5.5	43.27 ± 5.95	26.67
$P=0.001$

Figure 3. Correlation of MDR1-3435 genotype with the dose-adjusted trough concentration of cyclosporine in patients who co-administered with diltiazem. Dose-adjusted trough concentration of cyclosporine was significantly higher in MDR1-3435TT carriers than that in CC plus CT carriers.

group, the adjusted CsA trough concentrations in carriers of haplotypes T-T-T and T-T-C were significantly higher than in non-carriers ($P=0.009$ and 0.004 , respectively), but haplotype T-G-T carriers had significantly lower adjusted CsA trough concentrations than non-carriers ($P=0.002$).

Discussion

This study extensively investigated the effect of diltiazem on the relationship between genetic polymorphisms of MDR1 and CsA concentration during the stable stage after renal transplantation. In recent years, high inter-individual heterogeneity in the MDR1 gene, influencing the metabolism of

digoxin^[15–17], cyclosporin^[18–20], tacrolimus^[21–23] and amlodipine^[24], has been described. In the present study, we explored the association of MDR1 SNPs with CsA dose requirements and adjusted concentration in renal recipients co-treated or not with diltiazem. The ultimate objective of the present study was to optimize the clinical CsA therapeutic regimen, which might lead to individualized drug dosing and improved therapeutics.

In the present study, MDR1 C1236T did not influence the adjusted CsA trough concentration during the stable stage in all patients; however, MDR1 C3435T influenced the adjusted CsA trough concentration in patients using diltiazem. These results suggest that individuals carrying the MDR1-2677TT genotype or the T-T-T or T-T-C MDR1 haplotypes have a higher adjusted CsA trough concentration, regardless of co-treatment or not with diltiazem. Moreover, this result also suggests that the CsA dosage for patients with these genotypes and haplotypes might be reduced during the stable stage after renal transplantation.

The MDR1 variant alleles, 1236-T and 2677-T, significantly lower P-gp mRNA expression compared with 1236-C and 2677-G, respectively^[7]. Dennis *et al*^[18] reported that MDR1 C3435T did not influence the dose-adjusted trough blood concentration of CsA in stable renal transplant patients. Consistent results were obtained in another study involving American renal transplant patients^[19]. Crettol *et al*^[20] also reported that MDR1 genotypes did not influence the dose-adjusted trough blood concentration of CsA in transplant recipients. In contrast, Chinese renal transplant patients showed that MDR1 G2677T/A and MDR1 haplotypes C-G-C, T-G-T and T-T-C are associated with the CsA concentration during the early post-

Table 6. Difference of CsA dose, CsA concentration and adjusted CsA blood concentration among MDR1-3435 genotype in Dil(+) patients or Dil(-) patients.

Dil(+) (n=76)	MDR1-3435		
	CC	CT	TT
Patients number, <i>n</i>	40	2	9
CsA daily dose ($\text{mg}\cdot\text{kg}^{-1}\cdot\text{d}^{-1}$)	2.79 ± 0.84	2.97 ± 0.56 $P=0.003$	2.07 ± 0.44
CsA trough concentration (ng/mL)	122.83 ± 30.15	136.12 ± 26.02 $P=0.077$	119.99 ± 27.23
Dose adjusted concentration (ng/mL per mg/kg)	45.18 ± 12.35	46.14 ± 7.55 $P=0.011$	58.83 ± 13.95
Dil(-) (n=50)	MDR1-3435		
	CC	CT	TT
Patients number, <i>n</i>	20	23	7
CsA daily dose ($\text{mg}\cdot\text{kg}^{-1}\cdot\text{d}^{-1}$)	3.78 ± 0.97	3.59 ± 0.97 $P=0.247$	3.18 ± 0.67
CsA trough concentration (ng/mL)	123.36 ± 28.69	103.67 ± 21.14 $P=0.094$	109.00 ± 16.64
Dose adjusted concentration (ng/mL per mg/kg)	33.47 ± 7.69	31.94 ± 10.44 $P=0.805$	33.77 ± 10.27

Table 7. Difference of adjusted CsA concentration between different haplotype in Dil(+) patients or Dil(-) patients.

Haplotype	Group	Dil(+), n=76 n	Adjusted concentration (ng/mL per mg/kg)	Group	Dil(-), n=50 n	Adjusted concentration (ng/mL per mg/kg)
T-G-C	Noncarriers	38	51.97±11.35	Noncarriers	27	32.36±8.35
	carriers	38	42.71±9.61 <i>P</i> =0.0001	carriers	23	32.57±6.36 <i>P</i> =0.991
C-G-C	Noncarriers	43	48.31±13.26	Noncarriers	30	33.83±7.79
	carriers	33	45.88±8.76 <i>P</i> =0.470	carriers	20	31.89±7.95 <i>P</i> =0.350
T-T-T	Noncarriers	52	44.82±11.21	Noncarriers	34	30.28±6.83
	carriers	24	52.47±10.17 <i>P</i> =0.007	carriers	16	36.82±7.78 <i>P</i> =0.009
T-G-T	Noncarriers	66	48.05±11.67	Noncarriers	35	34.73±7.45
	carriers	10	42.89±9.13 <i>P</i> =0.142	carriers	15	27.43±3.57 <i>P</i> =0.002
T-T-C	Noncarriers	69	45.23±10.52	Noncarriers	45	31.87±7.42
	carriers	7	64.57±9.25 <i>P</i> =0.001	carriers	5	41.89±5.18 <i>P</i> =0.004

transplant period^[25]. Consistently, Chen *et al*^[26] also reported that MDR1 SNPs and haplotypes were associated with C(2) and C(0) of CsA in 115 Chinese patients at 1 week and 1 month after renal transplantation. The MDR1 2677G allele has also been associated with a high CsA dose requirement to prevent renal allograft rejection in North India patients^[27].

In the present study, we observed a relationship between the MDR1C1236T or the MDR1G2677T polymorphism and the adjusted CsA concentration in Chinese renal recipients, regardless of diltiazem use during the stable stage. This result indicated a positive correlation between the number of MDR12677T alleles and adjusted CsA trough blood concentrations, showing that every T allele was associated with an approximate 20% increment in adjusted trough blood concentrations of CsA.

However, considering the relationship between MDR1 C3435T and the CsA concentration, we only observed a significant association in patients using diltiazem. Among the 50 SNPs of the MDR1 gene, the mutation at position 3435 in exon 26 is the only silent polymorphism identified to date that might influence P-gp expression in different human tissues and different ethnic groups^[28]. Previous studies have reported similar results, showing that MDR1 C3435T did not influence the adjusted CsA trough blood concentration in patients who did not use diltiazem during the early and stable post-transplant periods^[18, 20, 25]. We cannot explain this change in relationship; however, we speculate that this change might partially reflect the nature of diltiazem, which is a substrate and inhibitor of P-gp.

In the present study, the whole blood CsA concentration was measured using the FPIA method. It has previously been reported that most of the analytical methods were specific for

the parent drug, although some discrepancies in the results were obtained between high-performance liquid chromatography and fluorescence polarization immunoassays (FPIA). This overestimation might reflect cross-reactivity with CsA metabolites, even when monoclonal antibodies are used in the immunoassays^[8, 11, 28]. In the present study, the co-administration of diltiazem affected the correlation between MDR1 genetic polymorphisms and CsA blood concentrations, likely reflecting the reaction of FPIA with CsA and its metabolites. Thus, further mechanistic studies are needed to explain these findings.

In the present study, we investigated the impact of MDR1 haplotypes derived from SNPs C1236T, G2677T and C3435T on the adjusted CsA trough concentration in renal transplant patients and observed that the adjusted CsA concentration in carriers of haplotypes T-T-T and T-T-C was significantly higher than in non-carriers. Chowbaya *et al*^[29] reported that CsA exposure ($AUC_{0-4\text{ h}}$, $AUC_{0-12\text{ h}}$ and C_{max}) was higher in patients with the T-T-T haplotype than in heart transplant patients with the C-G-C haplotype. However, Ingrid *et al*^[30] showed that MDR1 haplotypes derived from the SNPs G2677T (exon 21) and C3435CT (exon 26) are not associated with cyclosporine pharmacokinetics in renal transplant patients. Therefore, prospective studies using a large sample size might be needed to explore the impact of MDR1 haplotypes on the CsA adjusted concentration.

In conclusion, the results of the present study demonstrated that the adjusted CsA trough concentration was significantly higher in MDR1-3435TT carriers than that in CC and CT carriers among patients using diltiazem; however, these differences were not observed in patients who did not use diltiazem. We also observed that G2677T/A SNPs and T-T-T and T-T-C haplotypes in MDR1 are associated with higher CsA trough

concentrations in Chinese renal transplant patients, regardless of the co-administration or not of diltiazem during the stable post-transplant period. The identification of MDR1 SNPs and haplotypes and the use of diltiazem might have a clinically significant impact on allograft outcome and might provide pre-transplant pharmacogenetics information to individualize the CsA dosage.

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Author contribution

Yi-xi WANG, Jia-li LI, Min HUANG, and Chang-xi WANG designed the study; Yi-xi WANG, Jia-li LI, and Yu ZHANG performed the experiments; Yi-xi WANG, Jia-li LI, and Xue-ding WANG contributed new reagents or analytic tools; Yi-xi WANG and Jia-li LI analyzed the data; and Yi-xi WANG and Jia-li LI drafted the manuscript.

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