INFLUENCE OF *ABCB-1* C3435T POLYMORPHISMS ON PLASMA NEVIRAPINE AND EFAVIRENZ LEVELS AND THEIR EFFECTS ON VIROLOGIC AND IMMUNOLOGICAL OUTCOMES IN HIV/TB CO-INFECTED THAI ADULTS UNDER ANTI-RETROVIRAL THERAPY

Sumonmal Uttayamakul[1,2], Sirirat Likanonsakul[2], Weerawat Manosuthi[2], Nuanjun Wichukchinda[3], Tatsuo Shioda[4] and Srisin Khusmith[1]

[1]Department of Microbiology and Immunology, Faculty of Tropical Medicine, Mahidol University, Bangkok; [2]Bamrasnaradura Infectious Diseases Institute, Department of Disease Control, Ministry of Public Health, Nonthaburi; [3]National Institute of Health, Department of Medical Sciences, Ministry of Public Health, Nonthaburi, Thailand; [4]Research Institute of Microbial Disease, Osaka University, Osaka, Japan

Abstract. ATP-binding cassette, sub-family B (encoded by *ABCB-1*or *MDR-1*) has an important role in cellular export of antiretroviral agents. A previous study showed that *ABCB-1* C3435T polymorphism affects plasma efavirenz and nelfinavir concentrations and rate of CD4+ T cell recovery after starting anti-retroviral treatment (ART). The present study examined the influence of *ABCB-1* polymorphisms on plasma nevirapine and efavirenz levels when co-administered with rifampicin in 124 HIV/TB patients who received nevirapine- (400 mg/day) (n = 59) and efavirenz- (600 mg/day) (n = 65) based ART. *ABCB-1* C3435T polymorphisms were genotyped using real-time PCR. CD4 T cell counts and HIV-1 viral RNA were evaluated in response to ART. The frequencies of CC, CT and TT genotypes of *ABCB-1* C3435T polymorphism were 34% (n = 42), 55% (n = 68) and 12% (n = 14), respectively. Contrary to the previous report, no association was found among these genotypes and plasma drug concentrations at weeks 6 and 12 of ART and after rifampicin discontinuation. We also observed no differences in CD4+ T cell recovery rate among different *ABCB-1* C3435T genotypes. In nevirapine group, however, all the patients with CT genotype achieved HIV-1 RNA levels of < 50 copies/ml, while 67% of those with TT and 95% with CC genotypes achieved < 50 copies/ml (p = 0.040). These data suggested that *ABCB-1* C3435T polymorphisms do not affect plasma nevirapine and efavirenz concentrations in HIV/TB co-infected Thai patients or their immunological outcome, but had an effect on virologic outcome in the nevirapine-treated group.

Keywords: *MDR-1 (ABCB-1*), HIV, tuberculosis, nevirapine, efavirenz

Correspondence: Dr Srisin Khusmith, Department of Microbiology and Immunology, Faculty of Tropical Medicine, Mahidol University, 420/6 Ratchawithi Road, Bangkok 10400, Thailand.
Tel: 66 (0) 2354 9100; Fax: 66 (0) 2643 5583
E-mail: tmskm@mahidol.ac.th

INTRODUCTION

Nevirapine- and efavirenz- based highly active antiretroviral therapy (HAART) have been mostly recommended regimen as components of the

first-line drugs for HIV-1 treatment worldwide (Hammer *et al*, 2006). These drugs are potent and preferable options for initial antiretroviral treatments (ART) in both HIV and HIV/tuberculosis (TB) co-infection. Concomitant administration of HAART and anti-TB medications can result in significant variations in drug exposure that may cause loss of efficacy or development of resistance, intolerance or toxicity (Bonora and Di Perri, 2008). Rifampicin is not only a critical component of TB therapy, but also a potent inducer of cytochrome P450 (CYP) enzyme activity (Ward *et al*, 2003). CYP450 and multidrug-resistance transporter gene 1 (*MDR-1* or recently named; ATP-binding cassette, sub-family B (*ABCB-1*) have been reported to play an important role in metabolism and transport of antiretroviral agents (Tsuchiya *et al,* 2004; Saitoh *et al,* 2005).

Although nevirapine- or efavirenz-based HAART has been used as the main therapy in Thailand, information regarding the influence of host genetic polymorphism on these drug levels among Thai populations is limited so far. We have recently reported the effects of *CYP2B6* G516T polymorphisms on plasma efavirenz and nevirapine levels when co-administered with rifampicin in HIV/TB co-infected Thai adults, in which *CYP2B6*-TT genotype has significant impact on plasma drug concentrations, while rifampicin co-administration has no effect (Uttayamakul *et al,* 2010). However, the effect of *ABCB-1* C3435T polymorphism on antiretroviral treatment response in HIV/TB co-infected Thais has never been evaluated.

MDR-1 or *ABCB-1* encodes P-glycoprotein (P-gp), which is an integral membrane protein of 170 kDa and is responsible for multidrug resistance in cancer cells and functions as an energy-dependent drug-transport pump (Kimchi-Sarfaty *et al,* 2007). P-gp actively pumps a variety of drugs, chemotherapeutic agents, cardiac drugs, antibiotics, steroids and antiretroviral drugs (Sparreboom *et al,* 2003; Sakaeda 2005). Several studies have suggested that *ABCB-1* C3435T mutant can predict more favorable virologic responses in patients with efavirenz-containing regimens (Fellay *et al,* 2002; Haas *et al,* 2005). This C3435T polymorphism is known to affect both single-dose and steady-state pharmacokinetic of efavirenz (Mukonzo *et al,* 2009). However, no influence on plasma drug levels was found in other studies (Winzer *et al,* 2003; Haas *et al,* 2004; Tsuchiya *et al,* 2004). *MDR-1* 3435T allele has been reported to delay disease progression to pediatric AIDS, but has no effect on HIV-1 vertical transmission (Bellusci *et al,* 2010), while it does not influence the response to antiretroviral therapy in drug naive HIV-positive patients (Benish *et al,* 2010). However, the findings from one population may not be generalized to other populations due to the ethnic differences in drug effect and body weight of the patients. High body weight was evidenced to be associated with low plasma concentration at weeks 6 and 12 of efavirenz-based ART in Thai HIV/TB patients (Manosuthi *et al,* 2009b).

Therefore, the main objective of the study was to investigate whether *ABCB-1* C3435T polymorphisms could influence plasma nevirapine and efavirenz levels when co-administered with rifampicin in HIV/TB infected Thai adults. HIV-1 RNA levels and CD4 T cell counts in relation to this polymorphism were used to evaluate their effects on virologic and immunological outcomes.

MATERIALS AND METHODS

Patients

One hundred twenty-four rifampicin recipients with concurrent HIV-1/TB coinfection were studied. Sixty-five patients received efavirenz- (600 mg/day) based ART, while 59 received nevirapine- (400 mg/day) based ART. These patients were initially recruited in a randomized control trial study to compare the efficacy of efavirenz and nevirapine among HIV-infected patients receiving rifampicin at Bamrasnaradura Infectious Diseases Institute (BIDI), Nonthaburi, Thailand since December 2006 (Manosuthi *et al*, 2009a). They are ARV naïve patients with active tuberculosis and received rifampicin-containing anti-TB regimen for 4-6 weeks prior to enrollment. In this study, only 124 patients who have complete data sets of plasma drug levels at week 6 and 12 of ART and 1 month after rifampicin discontinuation were included as previously described (Uttayamakul *et al*, 2010). The patients received oral lamivudine (150 mg) and stavudine (30 mg for those who weighed ≤60 kg and 40 mg for those who weighed >60 kg) every 12 hours. They were randomized to receive either efavirenz (600 mg) at bedtime while fasting or nevirapine (200 mg) every 12 hours after 2 weeks at a starting dose of 200 mg every 24 hours. The dosage of rifampicin was 450 mg/day for patients who weighed ≤50 kg and 600 mg/day for those who weighed >50 kg. The anti-TB drug regimen was isoniazid, rifampicin, ethambutol and pyrazinamide for the first two months, followed by isoniazid and rifampicin for the subsequent 4-7 months. The study was approved by Institutional Ethics Committees of Bamrasnaradura Infectious Diseases Institute (BIDI) and the Ministry of Public Health, Thailand and written informed consents were obtained from all participants.

Blood samples

EDTA blood samples were collected for single nucleotide polymorphism (SNP) genotyping, CD4 T cell counts and HIV-1 viral load. Lithium heparinized blood samples were collected after 12 hours of drug administration (C_{12}) at weeks 6 and 12 of ART and after rifampicin discontinuation for 1 month for analysis of plasma nevirapine and efavirenz concentrations. Plasma were separated by centrifugation at 1,800*g* for 20 minutes and stored at -20°C.

SNP genotyping of *ABCB-1* C3435T

Genomic DNA was extracted using QIAamp DNA blood Mini kit (QIAGEN, Hilden, Germany) and stored at -20°C until used for SNP genotyping. Genotyping of *ABCB-1* C3435T (rs 1045642) was carried out using real-time polymerase chain reaction (PCR) with allele-specific fluorogenic 5′ nuclease chain reaction assay of ABI PRISM 7500 sequence detection system (Applied Biosystems, Foster City, CA) as the primers and TaqMan MGB probe assay protocol ID C_7586657. Each 25 µl PCR mixture contained 20 ng of genomic DNA, 900 nM of primers, 200 nM TaqMan of minor groove binder (MGB) probe and 12.5 µl of TaqMan universal PCR master mix. The thermal cycler program was set up as follows: at 95°C for 10 minutes, repeated 45 cycles at 95°C for 15 seconds and 60°C for 1 minute. The plate was read using the allelic discrimination settings. The SNP assay was set up using SDS, version 1.3.0 as an absolute quantification assay. Post-assay analysis was performed using SDS software.

Determination of plasma nevirapine and efavirenz concentration

Plasma nevirapine and efavirenz

concentrations were measured using reverse phase high performance liquid chromatography (HPLC) method at the HIV-Netherlands-Australia-Thailand (HIV-NAT) Research Pharmacokinetic Laboratory, Chulalongkorn Medical Research Center, Bangkok, Thailand. HPLC was performed in accordance with the protocol developed by Department of Clinical Pharmacology, University Medical Center Nijmegan, Nijmegan, The Netherlands (Hollanders *et al,* 2000).

CD4 T lymphocyte counts and plasma HIV-1 RNA quantitation

CD4 T lymphocyte counts were analysed at baseline and every 12 weeks after initiation of antiretroviral treatment by flow cytometry using monoclonal antibodies with three colors reagent (TriTEST, Becton Dickinson BioSciences, Franklin Lakes, NJ) and analyzed by FACScan flow cytometer (Becton Dickinson BioSciences, San Jose, CA). Plasma HIV-1 RNA was determined by reverse transcriptase-PCR at baseline and every 12 weeks after initiation of ART and quantified using COBAS Amplicor, version 1.5 (Roche Molecular System Pleasanton, CA). The lower detection limit for HIV-1 RNA level is 50 copies/ml.

Statistical analysis

Different genotypes in relation to body weight, age at baseline and plasma drug levels were analysed using SPSS software version 14.0 (SPSS, Chicago, IL). One-way analysis of variance (ANOVA) was used to determine statistical significance levels for differences in mean values of plasma drug levels among various genotype groups. CD4 T cell counts and HIV-1 viral load in patients carrying different genotypes were compared by Kruskal-Wallis test. A difference in proportion of patients who achieved plasma HIV-1 RNA < 50 copies/ml at week 24 of ART was evaluated by chi-square or Fisher's exact test. A p-value of < 0.05 is considered statistically significant.

RESULTS

Patients' characteristics

The baseline characteristics of patients were evaluated for gender, age, body weight and liver enzymes. All 124 patients were ethnically Thai and among these, 68% and 65% were male in nevirapine and efavirenz group, respectively. The patients had mean age of 38.0±8.6 and 35.9±8.2 years and mean body weight of 54.4±9.4 kg and 53.3±9.8 kg in nevirapine and efavirenz group, respectively. When subgroups of bodyweight were analyzed at baseline in nevirapine group, the difference was seen in *ABCB-1* C3435 CT heterozygote (51.6±9.3) compared to CC (58.8±7.4) and TT genotypes (56.0±13.1) ($p = 0.018$), whereas no difference among patients receiving efavirenz with CT (54.9±11.7), CC (51.5±6.8) and TT genotypes (51.9±8.1) (p=0.540) was found. Similar levels of clinical laboratory parameters including alkaline phosphatase, aspartate aminotransferase and alanine aminotransferase were seen in both patient groups.

Frequencies of *ABCB-1* C3435T genetic polymorphisms

When the *ABCB-1* C3435T in 124 HIV/TB co-infected adults were genotyped, the frequency of CC, CT and TT genotypes was 34% ($n = 42$), 55% ($n = 68$) and 11% ($n = 14$), respectively. Likewise, the frequency in minor T allele genotype was 0.35 and 0.42 in nevirapine and efavirenz group, respectively. The overall minor allele frequency of *ABCB-1* C3435T polymorphisms in 124 cases was 0.39, which

Table 1
Genotype distribution and allele frequency of *ABCB-1* C3435T in HIV/TB co-infected Thai adults.

ARV treatment	*ABCB-1* C3435T genotype frequency (%)			No. of patients
	Wild type CC Number (%)	Heterozygous CT Number (%)	Homozygous TT Number (%)	
Nevirapine	21 (36)	35 (59)	3 (5)	59
Efavirenz	21 (32)	33 (51)	11 (17)	65
Overall genotype frequency	42 (34)	68 (55)	14 (11)	124
C allele frequency (%)	61			
T allele frequency (%)	39			

ABCB-1 C3435T polymorphisms were found to be in Hardy-Weinberg equilibrium.

was in agreement with Hardy-Weinberg equilibrium (Table 1).

Association of *ABCB-1* C3435T genetic polymorphisms with plasma nevirapine and efavirenz concentrations

The *ABCB-1* C3435T polymorphism was evaluated in relation to plasma nevirapine and efavirenz levels which showed that the mean plasma nevirapine concentrations of patients with CT genotype at weeks 6 and 12 of ART and one month after rifampicin discontinuation (6.0±4.2, 5.5±2.9 and 7.0±3.9 mg/l, respectively) were not significantly different from those with CC genotype (5.6±2.7, 5.6±2.3 and 6.9±2.4 mg/l, respectively) or TT genotype (5.4±1.6, 5.9±1.9 and 4±1.3 mg/l, respectively) (Table 2). Similarly, in efavirenz group, the mean plasma drug concentrations of patients with CT genotype at weeks 6 and 12 of ART and 1 month after rifampicin discontinuation (4.8±5.0, 5.7±7.8 and 3.7±3.2 mg/l, respectively) were not different from those with CC genotype (3.7±2.4, 3.0±2.7 and 3.5±2.1 mg/l, respectively) or TT genotype (3.6±2.0, 3.4±2.3 and 2.8±1.8 mg/l, respectively). It should be noted that rifampicin induces P-gp in hepatic and intestinal cells through activation of the nuclear receptors (Manceau *et al*, 2010), but its effect on plasma drug level during co-administration and after discontinuation could not be observed in this study.

Association of CD4 T cell counts with HIV-1 viral load among patients with *ABCB-1* C3435T genotypes

The numbers of CD4 T cells in patients with CC, CT and TT genotypes increased in a similar manner at weeks 12, 24, 36 and 48 of ART compared to the baseline in both nevirapine and efavirenz groups (Fig 1). No significant differences in the median CD4 T cell counts of each genotype at different time points were also seen in nevirapine and efavirenz groups (p=0.155, 0.582, 0.232, 0.756 and 0.573, respectively). When the number of CD4 T cell counts at week 24 of nevirapine based ART was compared to those at the baseline, patients with CT, CC, and TT genotypes gained 138 cells/µl (from

Table 2
Plasma nevirapine and efavirenz concentrations at weeks 6, 12 and after rifampicin discontinuation in each *ABCB-1* C3435T genotype.

Plasma drugs concentration	*N*	Mean ± SD of plasma efavirenz and nevirapine concentrations in each genotype			*p*-value
		CC	CT	TT	
Nevirapine (mg/l)	59	*n*=21	*n*=35	*n*=3	
Week 6[a]		5.6±2.7	6.0±4.2	5.4±1.6	0.872
Week 12[a]		5.6±2.3	5.5±2.9	5.9±2	0.972
After rifampicin discontinuation		6.9±2.4	7.0±3.9	4±1.3	0.330
Efavirenz (mg/l)	65	*n*=21	*n*=33	*n*=11	
Week 6[a]		3.7±2.4	4.8±5.0	3.6±2.0	0.523
Week 12[a]		3.0±2.7	5.7±7.8	3.4±2.3	0.234
After rifampicin discontinuation		3.5±2.1	3.7±3.2	2.8±1.8	0.663

[a]co-administration with rifampicin

38 to 176 cells/µl), 129 cells/µl (from 40 to 169 cells/µl), and 79 cells/µl (from 177 to 256 cells/µl), respectively, but there is no significant difference among these genotypes. In efavirenz group, patients with CT, CC, and TT genotypes gained 113 cells/µl (from 38 to 151 cells/µl), 138 cells/µl (from 57 to 195 cells/µl) 153 cells/µl (from 57 to 210 cells/µl), respectively. Again, there is no significant difference among these genotypes.

The proportions of patients with HIV-1 RNA levels less than 50 (log 1.69) copies/ml among the different *ABCB-1* C3435T genotypes at week 24 of ART were compared, which showed that in nevirapine group, 100% (34/34) of the patients with CT genotype achieved the HIV-1 RNA levels less than 50 copies/ml, while 67% (2/3) of those with TT genotype and 95% (20/21) of those with CC genotype achieved HIV-1 RNA levels less than 50 copies/ml (p = 0.040) (Fig 2). These proportions were paralleled the gain in CD4 cell counts at week 24 of ART from baseline. Conversely, in efavirenz group, 100% (11/11) of those with TT genotype, 97% (32/33) of those with CT genotype and 86% (18/21) of those with CC genotype achieved the HIV-1 RNA levels of less than 50 copies/ml. The difference in the proportion in efavirenz group, however, did not reach statistical significance. Consistently, there is a significant difference in HIV-1 RNA levels at 24 weeks of ART among CC, CT, and TT genotypes in nevirapine group (p = 0,012), but there is no such difference in efavirenz group.

DISCUSSION

In this study, the effects of *ABCB-1* C3435T polymorphism in HIV/TB co-infected Thai adults who received nevirapine- and efavirenz- based regimen were evaluated. This is the first report of the evaluation of the effects of *ABCB-1* C3435T gene polymorphisms on plasma nevirapine and efavirenz levels during rifampicin co-administration

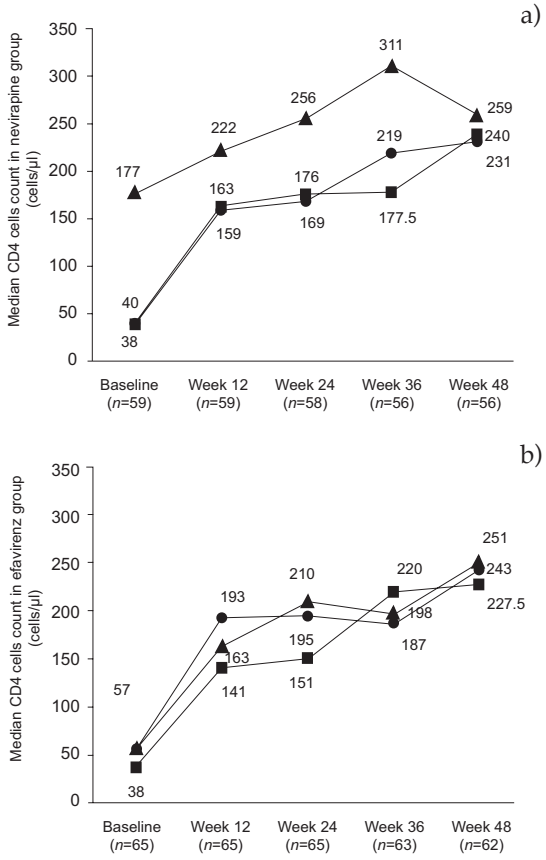

Fig 1–Time course of CD4 T cell counts among HIV/TB adults with different *ABCB-1* C3435T genotypes in (a) nevirapine group and (b) efavirenz group. (–●–) CC genotype, (–■–) CT genotype, (–▲–) TT genotype.

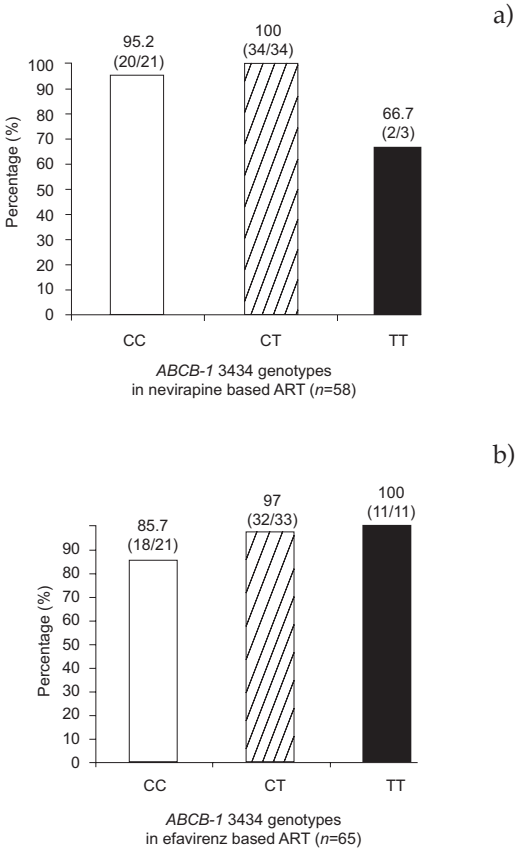

Fig 2–Percentage of patients reaching plasma HIV-1 RNA of less than 50 copies/ml at week 24 after starting anti-retroviral therapy among *ABCB-1* 3435-CC, CT and TT genotypes in (a) nevirapine group, p=0.040 by Fisher's exact test and (b) efavirenz group, p=0.229 by chi-square test.

and after discontinuation. Concerning the influence of *ABCB-1* C3435T polymorphism on plasma nevirapine levels, the results were concordance with the study in pregnant Thai women in which no association of this polymorphism on the persistence of plasma nevirapine concentration could be found (Chantarangsu *et al*, 2009). Likewise, no effect of *ABCB-1* C3435T polymorphism on plasma efavirenz levels was seen in this study as previously shown in several studies (Winzer *et al*, 2003; Haas *et al*, 2004; Tsuchiya *et al*, 2004) except for one study that showed significant differences among different *ABCB-1* C3435T genotype (Fellay *et al*, 2002).

The frequencies of *ABCB-1* 3435 CC, CT and TT among 124 Thai adults in the present study (33.9, 54.8 and 11.3%, re-

spectively) were slightly different from those Thai women in the recent report (32.4, 48.2 and 19.4%, respectively) (Chantarangsu *et al*, 2009). The CC genotype frequency among Thai populations in this study was higher than that reported among Caucasians (26%) but lower than West Africans (83%) and African Americans (61%) (Schaeffeler *et al*, 2001). It is also possible that ethnic diversity affects metabolism of antiretroviral drugs as Asian ethnicity is associated with a higher efavirenz accumulation in peripheral blood mononuclear cells (PBMCs) was evidenced (Elens *et al*, 2010).

In this study, *ABCB-1* C3435T polymorphism influenced differently on nevirapine- and efavirenz- based treatment outcome. Heterozygous CT genotype was associated with a more rapid virologic response in nevirapine-based antiretroviral therapy after 24 weeks, whereas patients on efavirenz-based regimen carrying homozygous TT showed a slightly better virologic response. The association between *ABCB-1* C3435T polymorphism and treatment outcome reported so far has been variable in many studies. In a previous retrospective study, patients with 3435TT genotype had more rapid CD4 recovery than those with CT and CC genotype after nelfinavir- or efavirenz-based treatment (Fellay *et al*, 2002). In other studies, however, no associations were found between *ABCB-1* C3435T polymorphism and either CD4 response or viral load (Nasi *et al*, 2003), and between genotype and either HIV-1 viral decay or CD4 response in an open label, single arm study (Fellay *et al*, 2002; Haas *et al*, 2003; Nasi *et al*, 2003). These discrepancies might be due to different study designs as clinical controlled trials would be the ideal context for such pharmacogenetic studies.

Although *ABCB-1* encodes P-gp and is responsible for drugs transport, nevirapine and efavirenz are not P-gp substrates (Stormer *et al*, 2002; Almond *et al*, 2005; Dirson *et al*, 2006). Nevertheless, efavirenz has been shown to induce P-gp function in an *in vitro* study (Weiss *et al*, 2009) and acts as an inducer of CYPs *in vivo* (Weiss *et al*, 2009). Therefore, several possible roles of P-gp in anti-retroviral drug metabolism have been proposed, including the interaction of P-gp with other cellular membrane proteins (Dirson *et al*, 2006). However, its function in other cell types is still unclear (Sankatsing *et al*, 2004). Furthermore, P-gp function on lymphocytes is affected by other factors including HIV infection, protease inhibitors and NNRTI drugs (Sankatsing *et al*, 2004). C3435T polymorphism has been found to modulate P-gp function in natural killer cells and the expression of *MDR1* mRNA in leukocytes (Hitzl *et al*, 2001). P-gp expression in the intestinal epithelial cells is also affected by *ABCB-1* C3435T polymorphisms (Hoffmeyer *et al*, 2000). The normal level of P-gp expression on C allele would lead to lower drug concentration in plasma compared to that in P-gp expression on T allele. However, controversial results were also reported (Sankatsing *et al*, 2004). Therefore, the effects of this polymorphism on the expression and function of P-gp in HIV patients receiving HAART needs to be further investigated. It is also important to examine the intracellular concentrations of anti-retroviral drugs in PBMCs to evaluate the treatment outcome, as the adverse events from efavirenz toxicity is correlated with drug concentrations in the intracellular compartment, but not with those in plasma (Clifford *et al*, 2005; Rotger *et al*, 2005).

It should be noted here that 9 efavirenz- and 16 nevirapine- treated patients

failed to continue the study. Reasons for the failure included development of adverse drug effects (such as hepatitis, skin rash), death, transfer to other hospitals, and loss to follow-up. Although the genotype distribution of *ABCB-1* C3435T in this study is in Hardy-Weinberg equilibrium, there was an apparent reduction of TT homozygotes in nevirapine group. It is possible that many of nevirapine patients who developed severe adverse drug effects were TT homozygotes. It is thus important to determine the *ABCB-1* C3435T genotypes of those who failed to continue the study.

In conclusion, the present results indicated that *ABCB-1* C3435T polymorphisms has no effect on plasma nevirapine or efavirenz concentration in HIV/TB co-infected Thai adults.

ACKNOWLEDGEMENTS

We thank the patients for their kind participation in the study, and the staff of Bamrasnaradura Infectious Diseases Institute (BIDI), Thailand for their help in obtaining patients' consents especially Ms Tippawan Rattanatham, Ms Samruay Nilkamhang, and Ms Supeda Tongyen. We also thank Dr Emi E Nakayama for critical reading of this manuscript. This study was supported by the Royal Golden Jubilee PhD Program (Grant No. PHD/0094/2551) of The Thailand Research Fund; Faculty of Tropical Medicine, Mahidol University; the Health Sciences Foundation and the Ministry of Education, Culture, Sports, Sciences and Technology of Japan and Bamrasnaradura Infectious Diseases Institute (BIDI), Thailand.

REFERENCES

Almond LM, Hoggard PG, Edirisinghe D, Khoo SH, Back DJ. Intracellular and plasma pharmacokinetics of efavirenz in HIV-infected individuals. *J Antimicrob Chemother* 2005; 56: 738-44.

Bellusci CP, Rocco CA, Aulicino PC, *et al.* MDR1 3435T and 1236T alleles delay disease progression to pediatric AIDS but have no effect on HIV-1 vertical transmission. *AIDS* 2010; 24: 833-40.

Benish RL, Rodriguez B, Zimmerman PA, Mehlotra RK. Comparative description of haplotype structure and genetic diversity of MDR1 (ABCB1) in HIV-positive and HIV-negative populations. *Infect Genet Evol* 2010; 10: 60-7.

Bonora S, Di Perri G. Interactions between antiretroviral agents and those used to treat tuberculosis. *Curr Opin HIV AIDS* 2008; 3: 306-12.

Chantarangsu S, Cressey TR, Mahasirimongkol S, *et al.* Influence of CYP2B6 polymorphisms on the persistence of plasma nevirapine concentrations following a single intra-partum dose for the prevention of mother to child transmission in HIV-infected Thai women. *J Antimicrob Chemother* 2009; 64: 1265-73.

Clifford DB, Evans S, Yang Y, *et al.* Impact of efavirenz on neuropsychological performance and symptoms in HIV-infected individuals. *Ann Intern Med* 2005; 143: 714-21.

Dirson G, Fernandez C, Hindlet P, *et al.* Efavirenz does not interact with the ABCB-1 transporter at the blood-brain barrier. *Pharm Res.* 2006; 23: 1525-32.

Elens L, Vandercam B, Yombi JC, Lison D, Wallemacq P, Haufroid V. Influence of host genetic factors on efavirenz plasma and intracellular pharmacokinetics in HIV-1-infected patients. *Pharmacogenomics* 2010; 11: 1223-34.

Fellay J, Marzolini C, Meaden ER, *et al.* Response to antiretroviral treatment in HIV-1-infected individuals with allelic variants of the multidrug resistance transporter 1: a pharmacogenetics study. *Lancet* 2002; 359: 30-6.

Haas DW, Ribaudo HJ, Kim RB, *et al.* Pharmacogenetics of efavirenz and central nervous system side effects: an Adult AIDS Clinical Trials Group study. *AIDS* 2004; 18: 2391-400.

Haas DW, Smeaton LM, Shafer RW, *et al.* Pharmacogenetics of long-term responses to antiretroviral regimens containing Efavirenz and/or Nelfinavir: an Adult AIDS Clinical Trials Group Study. *J Infect Dis* 2005; 192: 1931-42.

Haas DW, Wu H, Li H, *et al.* MDR1 gene polymorphisms and phase 1 viral decay during HIV-1 infection: an adult AIDS Clinical Trials Group study. *J Acquir Immune Defic Syndr* 2003; 34: 295-8.

Hammer SM, Saag MS, Schechter M, *et al.* Treatment for adult HIV infection: 2006 recommendations of the International AIDS Society-USA panel. *JAMA* 2006; 296: 827-43.

Hitzl M, Drescher S, van der Kuip H, *et al.* The C3435T mutation in the human MDR1 gene is associated with altered efflux of the P-glycoprotein substrate rhodamine 123 from CD56+ natural killer cells. *Pharmacogenetics* 2001; 11: 293-8.

Hoffmeyer S, Burk O, von Richter O, *et al.* Functional polymorphisms of the human multidrug-resistance gene: multiple sequence variations and correlation of one allele with P-glycoprotein expression and activity in vivo. *Proc Natl Acad Sci U S A* 2000; 97: 3473-8.

Hollanders RM, van Ewijk-Beneken Kolmer EW, Burger DM, Wuis EW, Koopmans PP, Hekster YA. Determination of nevirapine, an HIV-1 non-nucleoside reverse transcriptase inhibitor, in human plasma by reversed-phase high-performance liquid chromatography. *J Chromatogr B Biomed Sci Appl* 2000; 744: 65-71.

Kimchi-Sarfaty C, Oh JM, Kim IW, *et al.* A "silent" polymorphism in the MDR1 gene changes substrate specificity. *Science* 2007; 315: 525-8.

Manceau S, Giraud C, Decleves X, *et al.* Lack of P-glycoprotein induction by rifampicin and phenobarbital in human lymphocytes. *Int J Pharm* 2010; 395: 98-103.

Manosuthi W, Sungkanuparph S, Tantanathip P, *et al.* A randomized trial comparing plasma drug concentrations and efficacies between 2 nonnucleoside reverse-transcriptase inhibitor-based regimens in HIV-infected patients receiving rifampicin: the N2R Study. *Clin Infect Dis* 2009a; 48: 1752-9.

Manosuthi W, Sungkanuparph S, Tantanathip P, *et al.* Body weight cutoff for daily dosage of efavirenz and 60-week efficacy of efavirenz-based regimen in human immunodeficiency virus and tuberculosis coinfected patients receiving rifampin. *Antimicrob Agents Chemother* 2009b; 53: 4545-8.

Mukonzo JK, Roshammar D, Waako P, *et al.* A novel polymorphism in ABCB1 gene, CYP2B6*6 and sex predict single-dose efavirenz population pharmacokinetics in Ugandans. *Br J Clin Pharmacol* 2009; 68: 690-9.

Nasi M, Borghi V, Pinti M, *et al.* MDR1 C3435T genetic polymorphism does not influence the response to antiretroviral therapy in drug-naive HIV-positive patients. *AIDS* 2003; 17: 1696-8.

Rotger M, Colombo S, Furrer H, *et al,* Influence of CYP2B6 polymorphism on plasma and intracellular concentrations and toxicity of efavirenz and nevirapine in HIV-infected patients. *Pharmacogenet Genomics* 2005; 15: 1-5.

Saitoh A, Singh KK, Powell CA, *et al.* An MDR1-3435 variant is associated with higher plasma nelfinavir levels and more rapid virologic response in HIV-1 infected children. *AIDS* 2005; 19: 371-80.

Sakaeda T. MDR1 genotype-related pharmacokinetics: fact or fiction? *Drug Metab Pharmacokinet* 2005; 20: 391-414.

Sankatsing SU, Beijnen JH, Schinkel AH, Lange JM, Prins JM. P glycoprotein in human immunodeficiency virus type 1 infection

and therapy. *Antimicrob Agents Chemother* 2004; 48: 1073-81.

Schaeffeler E, Eichelbaum M, Brinkmann U, *et al.* Frequency of C3435T polymorphism of MDR1 gene in African people. *Lancet* 2001; 358: 383-4.

Sparreboom A, Danesi R, Ando Y, Chan J, Figg WD. Pharmacogenomics of ABC transporters and its role in cancer chemotherapy. *Drug Resist Updat* 2003; 6: 71-84.

Stormer E, von Moltke LL, Perloff MD, Greenblatt DJ. Differential modulation of P-glycoprotein expression and activity by non-nucleoside HIV-1 reverse transcriptase inhibitors in cell culture. *Pharm Res* 2002; 19: 1038-45.

Tsuchiya K, Gatanaga H, Tachikawa N, *et al.* Homozygous CYP2B6 *6 (Q172H and K262R) correlates with high plasma efavirenz concentrations in HIV-1 patients treated with standard efavirenz-containing regimens. *Biochem Biophys Res Commun* 2004; 319: 1322-6.

Uttayamakul S, Likanonsakul S, Manosuthi W, *et al.* Effects of CYP2B6 G516T polymorphisms on plasma efavirenz and nevirapine levels when co-administered with rifampicin in HIV/TB co-infected Thai adults. *AIDS Res Ther* 2010; 7: 8.

Ward BA, Gorski JC, Jones DR, Hall SD, Flockhart DA, Desta Z. The cytochrome P450 2B6 (CYP2B6) is the main catalyst of efavirenz primary and secondary metabolism: implication for HIV/AIDS therapy and utility of efavirenz as a substrate marker of CYP2B6 catalytic activity. *J Pharmacol Exp Ther* 2003; 306: 287-300.

Weiss J, Herzog M, Konig S, Storch CH, Ketabi-Kiyanvash N, Haefeli WE. Induction of multiple drug transporters by efavirenz. *J Pharmacol Sci* 2009; 109: 242-50.

Winzer R, Langmann P, Zilly M, *et al.* No influence of the P-glycoprotein genotype (MDR1 C3435T) on plasma levels of lopinavir and efavirenz during antiretroviral treatment. *Eur J Med Res* 2003; 8: 531-4.