The Impact of Cytochrome P450 3A5 Genotype on Early Tacrolimus Metabolism and Clinical Outcomes in Lung Transplant Recipients

Wenwen Du

China-Japan Friendship Hospital

Xiaoxing Wang

China-Japan Friendship Hospital

Dan Zhang

China-Japan Friendship Hospital

Wenqian Chen

China-Japan Friendship Hospital

Xianglin Zhang

China-Japan Friendship Hospital

Pengmei Li (✉ lipengmei@yeah.net)

China-Japan Friendship Hospital

Research Article

Keywords: Tacrolimus, Lung transplantation, CYP3A5 genotype, Mortality, AKI

Posted Date: August 10th, 2021

DOI: <https://doi.org/10.21203/rs.3.rs-782512/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License. [Read Full License](#)

Version of Record: A version of this preprint was published at International Journal of Clinical Pharmacy on December 3rd, 2021. See the published version at <https://doi.org/10.1007/s11096-021-01359-3>.

Abstract

Background Tacrolimus (Tac) is the cornerstone of immunosuppressant therapy after lung transplantation (LTx). It shows great inter-individual variability in pharmacokinetics, which could partly be explained by pharmacogenetic factors.

Objective We aim to investigate the effect of cytochrome P450 3A5 (*CYP3A5*) (rs776746) genotypes on early post-operative Tac metabolism and clinical outcomes in LTx recipients.

Methods 90 recipients who underwent LTx from 2017 to 2019 at our institution were enrolled in the study. The effect of *CYP3A5* genotype on Tac concentration, dose, dose adjusted concentration (C/D) and interaction with azole antifungals were assessed during week 1–4 after transplantation. Associations between *CYP3A5* genotype and the incidence of acute kidney injury (AKI), length of hospital stay and mortality were analyzed.

Results *CYP3A5**1 carriers had lower C/D than *CYP3A5**3/*3 group at all time points ($p < 0.05$). To reach comparable blood concentrations, *CYP3A5**1 carriers required higher doses compared with *CYP3A5**3/*3 group ($p < 0.05$). Use of azole antifungals did not blunt the effect of *CYP3A5* genotypes on Tac metabolism. Logistic regression showed Tac concentration at week 1, not *CYP3A5* genotype, was associated with the incidence of AKI. No statistically significant difference was found between *CYP3A5* genotypes and the length of hospital stay (48 (37–68) vs 46(32–57) days, $p = 0.264$). Kaplan–Meier analysis showed no statistically significant difference between 30-day or 1-year mortality and *CYP3A5* genotype.

Conclusion *CYP3A5* genotype could affect Tac metabolism early after LTx. However, it has no influence on the incidence of AKI, length of hospital stay and mortality.

Impacts On Practice

- Genetic polymorphism of *CYP3A5**3 could affect tacrolimus metabolism early after lung transplantation and hence may provide information in determining personalized dosing of tacrolimus.
- To achieve target concentration, *CYP3A5**3/*3 carriers require lower doses compared with *CYP3A5**1 carriers, regardless of use of azole antifungals or not.
- Genetic polymorphism of *CYP3A5**3 *does not seem to be* directly associated with incidence of AKI and mortality in lung transplant recipients.
- Rather than considering *CYP3A5**3 genotype, paying more attention to tacrolimus concentration at week 1 seems more important in determining patient at higher risk of AKI after lung transplantation.

Introduction

Tacrolimus (Tac) is the cornerstone of immunosuppressant therapy after lung transplantation (LTx) [1]. It is essential to reach target range of Tac concentration as soon as possible in the early post-transplant period, since erratic concentration may increase the incidence of rejection [2], nephrotoxicity [3] and even affects mortality [4]. However, this attempt is usually hampered by its highly variable pharmacokinetics [5]. Thus, extensive monitoring of whole blood concentration is required for dose adjustment, which consumes time and resources, and adds economic burden and physical pain to patients [6].

As a supplement to routine concentration monitoring, pharmacogenetics may provide guidance information for Tac dosage adjustment. Tac is mainly metabolized by cytochrome P450 (CYP) 3A5 after oral administration [7]. *CYP3A5**3 (rs776746) mutation results in non-functional protein, leading to decreased *CYP3A5* activity [8]. And hence, *CYP3A5**3/*3 genotype carriers are reported to require lower doses, and exhibit higher dose adjusted trough concentration (C/D) of Tac when compared to *CYP3A5**1 carriers in kidney [9], liver [10], and heart transplantation recipients [11]. In LTx, similar results have been reported in Caucasians [12–14]. But studies in Chinese population are limited till now [15, 16], and Chinese has a different mutation rates of *CYP3A5**3 from Caucasians [6], so, there is a major void in this research area.

Aim of the study

Our study was designed to investigate two key questions. First, whether *CYP3A5* genotype plays a role in inter-individual variability of Tac metabolism early after LTx. Second, whether there is association between *CYP3A5* genotype and clinical outcomes.

Ethics approval

All procedures in this study were in accordance with the 1964 Helsinki declaration and its amendments, and was approved by the Ethics Committee of China-Japan Friendship Hospital in May, 2019 (No. 2019-65-K45). All recipients signed informed consent when undergoing lung transplantation, in which they gave permission for use of anonymized medical data by scientists.

Methods

Patients

Recipients who undergone LTx during 2017 to 2019 in China-Japan Friendship Hospital were enrolled in this study. The inclusion criteria were: (1) LTx for the first time; (2) received Tac-based immunosuppressive regimen; (3) age ≥ 18 . The exclusion criteria were: (1) received cyclosporin in the first month; (2) survived less than 24 h after surgery.

Immunosuppressive and anti-fungal regimens

Per protocol in our center, most recipients used a triple maintenance immunosuppressive regimen consisting of Tac, mycophenolate mofetil (MMF) and prednisone. Tac was given on post-operative day (POD) 1. Starting dose was 2 mg/d in 2 divided doses, except those with exceptionally high or low body weight were given a personalized dose per attending physician's judgement. Dose adjustments were made according to whole-blood trough concentration, determined using micro-particle enzyme immunoassay (ARCHITECT i1000SR immunoassay analyzer, Abbott U.S.). The target range of concentration was 8–10 ng/mL in the first month after LTx. The initial dose of MMF was 1000 mg/d in 2 divided doses. Dose adjustments were made based on area under curve (AUC) calculations. Prednisone was given intravenously at 1 mg/kg/d during POD 1–3, and then switched to oral administration at 0.5 mg/kg/d, and gradually tapered to 5–10 mg/d for long-term maintenance.

In addition to immunosuppressive therapy, voriconazole or posaconazole were administrated for prophylaxis or treatment of invasive aspergillus. Voriconazole was started with a loading dose of 800 mg/d divided in 2 doses, either intravenous or oral. Then dose adjustments were made according to trough concentration measurements and maintenance dose was 300–400 mg/d. Posaconazole was given in suspensions, and started at 800 mg/d, in 4 divided doses. Then dose adjustments were made according to trough concentration and maintained at 600 or 800 mg/d, in 3 or 4 divided doses, respectively.

Genotype Determination

Genotypes of *CYP3A5**3 were determined using Sanger Sequencing. Briefly, genomic DNA was extracted from EDTA-anticoagulated peripheral whole blood samples by a commercially available DNA purification kit (EasyPure Blood Genomic DNA Kit, Transgene Biotech, Beijing, China). DNA samples were sequenced by the ABI 3730XL DNA Sequencer (ABI Co; Majorbio Biotechnology Co.,Ltd., Beijing, China).

Data collection

Patients' data were extracted by reviewing electronic medical records. Information including basic demographics (age, sex, height, weight, diagnosis, comorbidities, and transplant type), baseline laboratory test results (serum creatinine (SCr), anine aminotransferase (ALT), aspartate aminotransferase (AST), total bilirubin (Tbil), direct bilirubin (Dbil), urea, uric acid, red blood cells (RBC), hemoglobin, hematocrit, and glucose) and post-operative data (Tac dose (mg/d), corresponding body weight (kg), trough concentration (ng/mL), use of azole antifungals (voriconazole or posaconazole), and SCr) were obtained. Dose adjusted concentration (C/D, ng/mL per mg/kg/d) was calculated by dividing concentration by the corresponding body weight adjusted daily dose (mg/kg/d).

Outcome definitions

Pharmacokinetic outcomes of interest were Tac concentration, dose, and C/D. Clinical outcomes were incidence of AKI, length of hospital stay, 30-day and 1-year mortality. AKI was determined and staged according to the 'Kidney Disease: Improving Global Outcomes' (KDIGO) Clinical Practice Guideline, basing on SCr obtained from the transplantation day until 7 days after transplantation [17].

Statistical analysis

The normality distribution of data was assessed using the Shapiro–Wilk test. Chi-square test was performed to assess the deviation of gene frequencies from Hardy–Weinberg equilibrium. Descriptive statistics were expressed as mean $\pm$ standard deviation (SD) for normally distributed continuous variables or median (interquartile range, IQR) for non-normally distributed continuous variables. Categorical variables were presented as count (percentage). Comparisons between continuous variables were analyzed by Student's t test or Mann–Whitney U test. Pearson's chi-square test or Fisher's exact test was used to compare differences in categorical variables. Logistic regression was performed to study factors associated with incidence of AKI. Potential risk factors with a $p < 0.20$ in univariate analysis were incorporated into multivariate logistic regression, which adopts a forward LR strategy. 30-day and 1-year survival rates stratified by *CYP3A5* genotype was analyzed by Kaplan–Meier analysis and compared using the log–rank test. $P < 0.05$ was considered of statistical significance. Data was processed using Statistical Package for Social Science (SPSS) 19.0.

Results

Patients' characteristics

During 2017 and 2019, 150 patients received LTx. From this study population, 51 were excluded due to not available for genetic testing, 3 were excluded due to missing data, 3 were excluded due to use of cyclosporin, 2 were excluded for death within 24 hours and 1 was excluded due to less than 18 years old, leaving 90 for final analysis. Table 1 shows the demographics of the study population. Median (IQR) age was 60 (56–64) years old, and male patients (87.8%) accounted for a large proportion. Median (IQR) hospital stay length was 46 (35–60) days and follow-up time was 1 year.

For *CYP3A5* genotypes, **1/*1*, **1/*3* and **3/*3* were detected in 6, 36 and 48 patients, respectively. The frequency of *CYP3A5* genotypes were in accordance with Hardy–Weinberg equilibrium ($p > 0.05$). *CYP3A5*1* carriers and *CYP3A5*3/*3* group differ in baseline BMI, Dbil and pre-operative diabetes mellitus status.

Influence of *CYP3A5* genotype on Tac metabolism

Fig. 1a shows the Tac concentration trend-lines of all patients. Individual concentration fluctuated considerably in the early post-operative period, with only a small portion of patients reaching the target range on POD 3 after receiving a standardized starting dose.

*CYP3A5*1* carriers exhibited statistically significant lower concentration level than *CYP3A5*3/*3* group (3.8 (1.9–9.6) vs 8.8 (5.2–12.7) ng/mL, $p < 0.001$) (Fig. 1b) at comparable dose levels (2.0 (2.0–3.5) vs 2.0 (1.6–3.0) mg/d, $p = 0.221$) in week 1 (Fig. 1c). Lower C/D in *CYP3A5*1* carriers were observed at all time points: week 1 ($n = 81$), 80.58 (46.94–144.58) vs 232.27 (137.29–406.82) ng/mL per mg/kg/d, $p < 0.001$; week 2 ($n = 84$), 105.48 (63.91–219.46) vs 233.44 (131.70–521.40) ng/mL per mg/kg/d, $p < 0.05$; week 3 ($n = 86$), 89.15 (58.82–294.00) vs 212.52 (123.17–552.00) ng/mL per mg/kg/d, $p < 0.001$; and week 4 ($n = 81$), 105.30 (50.41–245.98) vs 229.26 (129.58–435.30) ng/mL per mg/kg/d, $p < 0.05$) (Fig. 1d).

Relationship between azole antifungals, *CYP3A5* genotype and Tac metabolism

Patients who underwent concomitant azole antifungals and Tac therapy showed significantly lower dose requirements and higher C/D of Tac at week 1–4 (Table 2) compared to those who received Tac only. When further stratified by *CYP3A5* genotype, *CYP3A5*1* carriers still showed lower C/D than *CYP3A5*3/*3* group at all time points (Fig. 2): week 1 without antifungals ($n = 70$), 72.45 (42.47–131.41) vs 210.06 (129.75–323.30) ng/mL per mg/kg/d, $p < 0.001$; week 1 with antifungals ($n = 11$), 417.60 (189.23–449.50) vs 970.70 (790.95–1311.10) ng/mL per mg/kg/d, $p < 0.05$; week 2 without antifungals ($n = 67$), 79.27 (61.08–184.27) vs 176.07 (110.55–266.98) ng/mL per mg/kg/d, $p < 0.05$; week 2 with antifungals ($n = 17$), 467.50 (220.80–745.20) vs 957.60 (696.48–1266.60) ng/mL per mg/kg/d, $p < 0.05$; week 3 without antifungals ($n = 63$), 61.50 (57.29–143.08) vs 132.00 (112.71–237.73) ng/mL per mg/kg/d, $p < 0.001$; week 3 with antifungals ($n = 23$), 320.17 (174.00–459.20) vs 881.87 (704.30–1154.25) ng/mL per mg/kg/d, $p < 0.001$; week 4 without antifungals ($n = 55$), 60.23 (46.84–149.24) vs 160.34 (95.36–246.67) ng/mL per mg/kg/d, $p < 0.001$; week 4 with antifungals ($n = 26$), 324.30 (207.80–543.60) vs 753.40 (435.30–961.13) ng/mL per mg/kg/d, $p < 0.05$).

Influence of *CYP3A5* genotype on AKI

Five variables, including age, baseline SCr, uric acid, RBC and week 1 Tac concentration ≥ 7.5 ng/mL, showed $p < 0.20$ in univariate analysis and were further assessed in multivariate model (Table 3). Only week 1 Tac concentrations ≥ 7.5 ng/mL (OR 5.38; 95% CI 1.40–20.69; $p = 0.014$) was associated with increased risk of AKI. *CYP3A5* genotype showed no significance in both univariate and multivariate analysis.

Influence of *CYP3A5* genotype on length of hospital stay and mortality

Of our study population, no statistically significant difference was observed in the length of hospital stay between *CYP3A5**1 carriers and *3/*3 group: 48 (37–68) vs 46(32–57) d, $p = 0.264$.

Thirty-day and 1-year survival rates of the study population were 94.4% and 88.9%, respectively. According to Kaplan-Meier analysis, although *CYP3A5**1 carriers showed higher survival rates than *3/*3 group at both 30-day (97.6% vs 91.7%) and 1-year (92.9% vs 85.4%), no statistical significance were achieved ($p = 0.367$ and $p = 0.433$, respectively) (Fig. 3).

Discussion

Statement of key findings

In this study, the influence of *CYP3A5* genotype on Tac metabolism and clinical outcomes in Chinese LTx population was discussed. We drew three major conclusions from the data: (1) patients bearing *CYP3A5**1 allele showed significantly lower Tac C/D and higher dose requirement compared with *3/*3 genotype in the early post-operative period; (2) the use of azole antifungals did not change the influence of *CYP3A5* genotype on Tac metabolism; (3) *CYP3A5* genotype has no effect on the incidence of AKI, length of hospital stay or mortality.

The frequency of *CYP3A5**3 variant differs among world populations, which is estimated to be 94, 68–71 and 18% in Europeans, Asians, and Africans, respectively [6]. The mutation rate in this study was 73%, comparable to previous studies in Chinese transplant recipients [16, 18].

A meta-analysis summarized that Tac C/D in kidney transplant recipients with *CYP3A5**3/*3 genotype was 1.8–2.5 times higher than *CYP3A5**1 carriers during the first year after transplantation [19]. Similar conclusions were also drawn in liver and heart transplant recipients [11, 20, 21]. Compared with other types of solid organ transplantation, studies concerning *CYP3A5* genotype in LTx population are limited [4, 13]. In Caucasians, Miano et al reported patients bearing *CYP3A5**3/*3 genotype exhibited approximately 50% higher C/D than *CYP3A5**1 carriers in the first 2 weeks after LTx [13] and Zheng et al investigated later time points, from 1 month to 12 months, and also observed higher C/D in *CYP3A5**3/*3 [4]. Only 1 study conducted in Chinese with a relatively small sample size of 51 recipients reported the C/D for *CYP3A5**3/*3 genotype was 1.9–2.5 times higher than *CYP3A5**1 carriers in the first month [16]. In our study, the C/D for *CYP3A5**3/*3 vs. *CYP3A5**1 carriers were 2.88, 2.21, 2.38 and 2.18 during week 1–4, respectively, similar to the results from both Caucasian and Chinese population.

Considering the difference in Tac-metabolizing activity, the 2015 Clinical Pharmacogenetics Implementation Consortium (CPIC) guideline and 2019 Therapeutic Drug Monitoring (TDM) consensus report have suggested using approximately 1.5–2 times higher starting dose for *CYP3A5**1 carriers in order to reach the same target concentration as *CYP3A5**3/*3 genotype [1, 6]. Due to the retrospective nature of our study, we could not control the initial dose of Tac, but, still, we found the dose requirement of *CYP3A5**1 carriers was 1.7–2.4 times higher than *CYP3A5**3/*3 when comparable concentrations were achieved (Fig. 1b and 1c). Therefore, genotyping of *CYP3A5* before Tac treatment may offer useful information in determining individualized dose [22].

Azole antifungals (voriconazole or posaconazole), often concomitantly used with Tac due to the high incidence of invasive aspergillosis after LTx, can increase Tac C/D by inhibiting CYP3A enzymes [14, 23]. Previous studies have reported a major drug-drug interaction between Tac and azole antifungals in both *CYP3A5**1 carriers and *CYP3A5**3/*3 group, and leads to significantly higher Tac C/D in lung and kidney transplant recipients [14, 24]. We also observed elevated Tac C/D in patients using azole antifungals. Moreover, *CYP3A5**3/*3 group still exhibited higher C/D and lower dose requirement compared with *CYP3A5**1 carriers (Fig. S1 and S2) regardless of using azole antifungals or not, suggesting the effect of *CYP3A5* genotype on Tac metabolism was not blunted by azole antifungals.

We further explored if a direct association exists between *CYP3A5* genotype and clinical outcomes. Since *CYP3A5* is expressed in the kidney, it is plausible to hypothesize that the genetic variability in *CYP3A5* expression and activity may affect the metabolism of Tac in kidney tubular cells, and therefore, may correlate with its nephrotoxicity [25, 26]. However, In analysis of risk factors of AKI, Tac concentration ≥ 7.5 ng/mL in the first week, not *CYP3A5* genotype, was associated with the incidence of AKI. Similarly, Woodahl et al reported no correlation between *CYP3A5* genotype and AKI in hematopoietic cell transplantation recipients [27]. Calabrese et al investigated the impact of *CYP3A5*, *CYP3A4*, and *ABCB1* genotypes on AKI in 321 LTx recipients and found no difference among different *CYP3A* genotypes [14]. One possible explanation for the lack of correlation is common genetic polymorphisms of *CYP3A5* did

not reflect its expression profile in kidney. The location and expression of renal *CYP3A5* might be the actual contributor to the nephrotoxicity of Tac [28].

CYP3A5 genotypes did not alter the length of hospital stay in LTx recipients. Previous study has reported increased hospital stay in kidney transplant recipients with *CYP2C19*2/*2* genotype [29]. Bosó et al explained the difference was directly correlated with higher median Tac C/D during the first week after transplant. Judging from our data, *CYP3A5*3/*3* group also exhibited higher median Tac C/D at week 1, however, no difference in hospital stay were found between genotypes. Considering the small number of patients (n=4 in *CYP2C19*2/*2* group) in Bosó's study, the correlation might lack potency and needs further validation. Furthermore, no correlation between *CYP3A5* genotype and mortality was found, which is in consistence with previous study in kidney and heart transplant population [11, 30].

Strengths And Weaknesses

In summary, despite the large number of studies addressing the influence of *CYP3A5* genotype on Tac metabolism, whether it plays a role in clinical outcomes remains controversial. Several meta-analysis and studies have reported no significant association between *CYP3A5* genotype and adverse outcomes [31, 32]. Besides, Tac C/D has been proposed as a stronger prognostic marker for poor outcomes recently. Though closely related to *CYP3A5* genotype, Tac C/D is also affected by other factors, including the use of corticosteroids and non-adherence, and therefore, may be better in reflecting immunosuppressive status [33, 34].

There are certain limitations in our study. First, this is a retrospective study with a relatively small sample size, therefore, the conclusions may need confirmation in prospective studies. Second, our population mainly consists of male adults, limited females and no pediatric patients were included. They may possess different physiological characteristics, such as liver function, hemoglobin concentration and hematocrits, and hence further studies in a more diverse population are warranted.

Conclusions

In this study, we investigated the influence of *CYP3A5* genotype on Tac metabolism and clinical outcomes in Chinese LTx recipients. As with other types of transplantation, *CYP3A5*1* carriers exhibited significantly lower C/D and higher dose requirements compared to *CYP3A5*3/*3* group in the early post-operative period. Concomitant use of azole antifungals did not blunt the influence of *CYP3A5* genotype on Tac metabolism. We also found *CYP3A5* genotype was not associated with clinical outcomes, including incidence of AKI, length of hospital stay, 30-day and 1-year mortality.

Declarations

Funding The study was financed with National Key Research and Development Program of China (No.2020YFC2005504). The funding source had no effect on any part of the study, preparation, or submission of the manuscript.

Conflicts of interest All authors report no conflict of interest.

Funding The study was financed with National Key Research and Development Program of China (No.2020YFC2005504). The funding source had no effect on any part of the study, preparation, or submission of the manuscript.

Conflicts of interest All authors report no conflict of interest.

Availability of data and material Upon reasonable request from the corresponding author.

Code availability Not applicable.

Authors' contributions DWW contributed to the design of the study. WXX collected the data. DWW and WXX analyzed the data. DWW, ZD and CWQ contributed to the preparation of the manuscript. ZXL and LPM revised the manuscript.

Ethics approval All procedures in this study were in accordance with the 1964 Helsinki declaration and its amendments, and was approved by the Ethics Committee of China-Japan Friendship Hospital (No. 2019-65-K45).

Consent to participate All recipients signed informed consent when undergoing Lung transplantation, in which they gave permission for use of anonymized medical data by scientists.

References

1. Birdwell KA, Decker B, Barbarino JM, Peterson JF, Stein CM, Sadee W, Wang D, Vinks AA, He Y, Swen JJ, Leeder JS, van Schaik R, Thummel KE, Klein TE, Caudle KE, MacPhee IA (2015) Clinical pharmacogenetics implementation consortium (CPIC) guidelines for CYP3A5 genotype and tacrolimus dosing. *Clin Pharmacol Ther* 98:19-24. <http://doi.org/10.1002/cpt.113>
2. Gueta I, Markovits N, Yarden-Bilavsky H, Raichlin E, Freimark D, Lavee J, Loebstein R, Peled Y (2018) High tacrolimus trough level variability is associated with rejections after heart transplant. *Am J Transplant* 18:2571-2578. <http://doi.org/10.1111/ajt.15016>
3. Sikma MA, Hunault CC, van de Graaf EA, Verhaar MC, Kesecioglu J, de Lange DW, Meulenbelt J (2017) High tacrolimus blood concentrations early after lung transplantation and the risk of kidney injury. *Eur J Clin Pharmacol* 73:573-580. <http://doi.org/10.1007/s00228-017-2204-8>
4. Zheng H, Zeevi A, Schuetz E, Lamba J, McCurry K, Griffith BP, Webber S, Ristich J, Dauber J, Iacono A, Grgurich W, Zaldonis D, McDade K, Zhang J, Burckart GJ (2004) Tacrolimus dosing in adult lung transplant patients is related to cytochrome P4503A5 gene polymorphism. *J Clin Pharmacol* 44:135-140. <http://doi.org/10.1177/0091270003262108>
5. Staatz CE, Tett SE (2004) Clinical pharmacokinetics and pharmacodynamics of tacrolimus in solid organ transplantation. *Clin Pharmacokinet* 43:623-653. <http://doi.org/10.2165/00003088-200443100-00001>
6. Brunet M, van Gelder T, Åsberg A, Haufroid V, Hesselink DA, Langman L, Lemaître F, Marquet P, Seger C, Shipkova M, Vinks A, Wallemacq P, Wieland E, Woillard JB, Barten MJ, Budde K, Colom H, Dieterlen MT, Elens L, Johnson-Davis KL, Kunicki PK, MacPhee I, Masuda S, Mathew BS, Millán O, Mizuno T, Moes DAR, Monchaud C, Noceti O, Pawinski T, Picard N, van Schaik R, Sommerer C, Vethe NT, de Winter B, Christians U, Bergan S (2019) Therapeutic drug monitoring of tacrolimus-personalized therapy: Second consensus report. *Ther Drug Monit* 41:261-307. <http://doi.org/10.1097/FTD.0000000000000640>
7. Hesselink DA, Bouamar R, Elens L, van Schaik RH, van Gelder T (2014) The role of pharmacogenetics in the disposition of and response to tacrolimus in solid organ transplantation. *Clin Pharmacokinet* 53:123-139. <http://doi.org/10.1007/s40262-013-0120-3>
8. Hustert E, Haberl M, Burk O, Wolbold R, He YQ, Klein K, Nuessler AC, Neuhaus P, Klattig J, Eiselt R, Koch I, Zibat A, Brockmöller J, Halpert JR, Zanger UM, Wojnowski L (2001) The genetic determinants of the CYP3A5 polymorphism. *Pharmacogenetics* 11:773-779. <http://doi.org/10.1097/00008571-200112000-00005>
9. Yaowakulpatana K, Vadcharavivad S, Ingsathit A, Areepium N, Kantachuvesiri S, Phakdeekitcharoen B, Sukasem C, Sra-lum S, Sumethkul V, Kitiyakara C (2016) Impact of CYP3A5 polymorphism on trough concentrations and outcomes of tacrolimus minimization during the early period after kidney transplantation. *Eur J Clin Pharmacol* 72:277-283. <http://doi.org/10.1007/s00228-015-1990-0>
10. Collier JK, Ramachandran J, John L, Tuke J, Wigg A, Doogue M (2019) The impact of liver transplant recipient and donor genetic variability on tacrolimus exposure and transplant outcome. *Br J Clin Pharmacol* 85:2170-2175. <http://doi.org/10.1111/bcp.14034>
11. Liu BY, Chen WQ, Chen ZG, Huang J, Liao ZK, Liu Q, Zheng Z, Song YH, Wang W, Hu SS (2019) The effects of CYP3A5 genetic polymorphisms on serum tacrolimus dose-adjusted concentrations and long-term prognosis in Chinese heart transplantation recipients. *Eur J Drug Metab Pharmacokinet* 44:771-776. <http://doi.org/10.1007/s13318-019-00563-x>
12. Monchaud C, de Winter BC, Knoop C, Estenne M, Reynaud-Gaubert M, Pison C, Stern M, Kessler R, Guillemain R, Marquet P, Rousseau A (2012) Population pharmacokinetic modelling and design of a bayesian estimator for therapeutic drug monitoring of tacrolimus in lung transplantation. *Clin Pharmacokinet* 51:175-186. <http://doi.org/10.2165/11594760-000000000-00000>
13. Miano TA, Flesch JD, Feng R, Forker CM, Brown M, Oyster M, Kalman L, Rushefski M, Cantu E, 3rd, Porteus M, Yang W, Localio AR, Diamond JM, Christie JD, Shashaty MGS (2020) Early tacrolimus concentrations after lung transplant are predicted by combined clinical and genetic factors and associated with acute kidney injury. *Clin Pharmacol Ther* 107:462-470. <http://doi.org/10.1002/cpt.1629>
14. Calabrese DR, Florez R, Dewey K, Hui C, Torgerson D, Chong T, Faust H, Rajalingam R, Hays SR, Golden JA, Kukreja J, Singer JP, Greenland JR (2018) Genotypes associated with tacrolimus pharmacokinetics impact clinical outcomes in lung transplant recipients. *Clin Transplant* 32:e13332. <http://doi.org/10.1111/ctr.13332>

15. Cai X, Song H, Jiao Z, Yang H, Zhu M, Wang C, Wei D, Shi L, Wu B, Chen J (2020) Population pharmacokinetics and dosing regimen optimization of tacrolimus in Chinese lung transplant recipients. *Eur J Pharm Sci* 152:105448. <http://doi.org/10.1016/j.ejps.2020.105448>
16. Zhang X, Xu J, Fan J, Zhang T, Li Y, Xie B, Zhang W, Lin S, Ye L, Liu Y, Jiang G (2017) Influence of IL-18 and IL-10 polymorphisms on tacrolimus elimination in Chinese lung transplant patients. *Dis Markers* 2017:7834035. <http://doi.org/10.1155/2017/7834035>
17. Khwaja A (2012) KDIGO clinical practice guidelines for acute kidney injury. *Nephron Clinical Practice* 120:c179-184. <http://doi.org/10.1159/000339789>
18. Tang HL, Xie HG, Yao Y, Hu YF (2011) Lower tacrolimus daily dose requirements and acute rejection rates in the CYP3A5 nonexpressers than expressers. *Pharmacogenet Genomics* 21:713-720. <http://doi.org/10.1097/FPC.0b013e32834a48ca>
19. Rojas L NI, Herrero MJ, Bosó V, Reig J, Poveda JL, Megías J, Bea S, Aliño SF (2015) Effect of CYP3A5*3 on kidney transplant recipients treated with tacrolimus: A systematic review and meta-analysis of observational studies. *Pharmacogenomics* 15:38-48. <http://doi.org/10.1038/tpj.2014.38>
20. Buendia JA, Bramuglia G, Staatz CE (2014) Effects of combinational CYP3A5 6986A>G polymorphism in graft liver and native intestine on the pharmacokinetics of tacrolimus in liver transplant patients: A meta-analysis. *Ther Drug Monit* 36:442-447. <http://doi.org/10.1097/ftd.0000000000000032>
21. Hendijani F, Azarpira N, Kaviani M (2018) Effect of CYP3A5*1 expression on tacrolimus required dose after liver transplantation: A systematic review and meta-analysis. *Clin Transplant* 32:e13306. <http://doi.org/10.1111/ctr.13306>
22. Andrews LM, Hesselink DA, van Schaik RHN, van Gelder T, de Fijter JW, Lloberas N, Elens L, Moes D, de Winter BCM (2019) A population pharmacokinetic model to predict the individual starting dose of tacrolimus in adult renal transplant recipients. *Br J Clin Pharmacol* 85:601-615. <http://doi.org/10.1111/bcp.13838>
23. Yamazaki H, Nakamoto M, Shimizu M, Murayama N, Niwa T (2010) Potential impact of cytochrome P450 3A5 in human liver on drug interactions with triazoles. *Br J Clin Pharmacol* 69:593-597. <http://doi.org/10.1111/j.1365-2125.2010.03656.x>
24. Vanhove T, Bouwsma H, Hilbrands L, Swen JJ, Spriet I, Annaert P, Vanaudenaerde B, Verleden G, Vos R, Kuypers DRJ (2017) Determinants of the magnitude of interaction between tacrolimus and voriconazole/posaconazole in solid organ recipients. *Am J Transplant* 17:2372-2380. <http://doi.org/10.1111/ajt.14232>
25. Kuehl P, Zhang J, Lin Y, Lamba J, Assem M, Schuetz J, Watkins PB, Daly A, Wrighton SA, Hall SD, Maurel P, Relling M, Brimer C, Yasuda K, Venkataramanan R, Strom S, Thummel K, Boguski MS, Schuetz E (2001) Sequence diversity in CYP3A promoters and characterization of the genetic basis of polymorphic CYP3A5 expression. *Nat Genet* 27:383-391. <http://doi.org/10.1038/86882>
26. Dai Y, Hebert MF, Isoherranen N, Davis CL, Marsh C, Shen DD, Thummel KE (2006) Effect of CYP3A5 polymorphism on tacrolimus metabolic clearance in vitro. *Drug Metab Dispos* 34:836-847. <http://doi.org/10.1124/dmd.105.008680>
27. Woodahl EL, Hingorani SR, Wang J, Guthrie KA, McDonald GB, Batchelder A, Li M, Schoch HG, McCune JS (2008) Pharmacogenomic associations in ABCB1 and CYP3A5 with acute kidney injury and chronic kidney disease after myeloablative hematopoietic cell transplantation. *Pharmacogenomics J* 8:248-255. <http://doi.org/10.1038/sj.tpj.6500472>
28. Metalidis C, Lerut E, Naesens M, Kuypers DR (2011) Expression of CYP3A5 and P-glycoprotein in renal allografts with histological signs of calcineurin inhibitor nephrotoxicity. *Transplantation* 91:1098-1102. <http://doi.org/10.1097/TP.0b013e328182177502>
29. Bosó V, Herrero MJ, Bea S, Galiana M, Marrero P, Marqués MR, Hernández J, Sánchez-Plumed J, Poveda JL, Aliño SF (2013) Increased hospital stay and allograft dysfunction in renal transplant recipients with Cyp2c19 AA variant in SNP rs4244285. *Drug Metab Dispos* 41:480-487. <http://doi.org/10.1124/dmd.112.047977>
30. Flahault A, Anglicheau D, Lorient MA, Thervet E, Pallet N (2017) Clinical impact of the CYP3A5 6986A>G allelic variant on kidney transplantation outcomes. *Pharmacogenomics* 18:165-173. <http://doi.org/10.2217/pgs-2016-0146>

31. Woillard JB, Gatault P, Picard N, Arnion H, Anglicheau D, Marquet P (2018) A donor and recipient candidate gene association study of allograft loss in renal transplant recipients receiving a tacrolimus-based regimen. Am J Transplant 18:2905-2913. <http://doi.org/10.1111/ajt.14894>

32. Khan AR, Raza A, Firasat S, Abid A (2020) CYP3A5 gene polymorphisms and their impact on dosage and trough concentration of tacrolimus among kidney transplant patients: A systematic review and meta-analysis. Pharmacogenomics J 20:553-562. <http://doi.org/10.1038/s41397-019-0144-7>

33. van Gelder T, Meziyerh S, Swen JJ, de Vries APJ, Moes D (2020) The clinical impact of the C₀/D ratio and the CYP3A5 genotype on outcome in tacrolimus treated kidney transplant recipients. Front Pharmacol 11:1142. <http://doi.org/10.3389/fphar.2020.01142>

34. Jouve T, Fonrose X, Noble J, Janbon B, Fiard G, Malvezzi P, Stanke-Labesque F, Rostaing L (2020) The TOMATO study (Tacrolimus Metabolization in Kidney Transplantation): Impact of the concentration-dose ratio on death-censored graft survival. Transplantation 104:1263-1271. <http://doi.org/10.1097/tp.0000000000002920>

Tables

Table 1 Baseline demographic characteristics of LTx patients stratified by *CYP3A5* genotype (n=90)

Parameters	All	<i>CYP3A5*1 carriers</i>	<i>CYP3A5*3/*3</i>	<i>p</i>
n (%)	90 (100.0)	42 (46.7)	48 (53.3)	
Age (years)	60 (56–64)	60 (53–64)	60 (56–64)	0.890
Male, n (%)	79 (87.8)	36 (85.7)	43 (89.6)	0.576
BMI (kg/m ²)	21.6±3.9	20.63±3.44	22.36±4.17	0.036
Diagnosis (n (%))				0.428
COPD	16 (17.8)	10 (23.8)	6 (12.5)	
ILD	60 (66.7)	27 (64.3)	33 (68.8)	
PH	10 (11.1)	3 (7.1)	7 (14.6)	
Others	4 (4.4)	2 (4.8)	2 (4.2)	
Comorbidities (n (%))				
Hypertension	16 (17.8)	4 (9.5)	12 (25.0)	0.055
Diabetes mellitus	30 (33.3)	9 (21.4)	21 (43.8)	0.025
Hyperlipidemia	7 (7.8)	3 (7.1)	4 (8.3)	0.833
Transplant type (n (%))				0.116
Unilateral	59 (65.6)	24 (57.1)	35 (72.9)	
Bilateral	31 (34.4)	18 (42.9)	13 (27.1)	
SCr (μmol/L)	53.8 (44.5–66.6)	51.3 (42.4–65.1)	56.0 (47.8–67.6)	0.070
ALT (IU/L)	18 (14–27)	20 (15–37)	17 (14–26)	0.486
AST (IU/L)	20 (16–23)	20 (17–23)	19 (15–24)	0.377
Tbil (μmol/L)	6.34 (4.74–10.20)	6.28 (4.63–11.44)	6.54 (4.89–9.74)	0.875
Dbil (μmol/L)	2.40 (1.70–3.06)	2.24 (1.63–2.80)	2.68 (1.88–3.72)	0.042
Urea (mmol/L)	6.22±1.86	6.08±1.73	6.33±1.97	0.531
Uric acid (μmol/L)	250 (194–338)	244 (186–291)	268 (200–355)	0.161
RBC (×10 ¹² cells/L)	4.50 (4.03–4.94)	4.52 (4.12–4.96)	4.48 (3.90–4.90)	0.428
Hemoglobin (g/L)	137 (125–148)	141 (126–149)	135 (125–147)	0.409
Hematocrit (%)	41.40 (37.68–44.55)	42.50 (37.95–45.32)	40.30 (37.38–43.85)	0.204
Glucose (mmol/L)	6.68 (5.19–8.22)	6.70 (5.05–8.26)	6.60 (5.65–8.21)	0.582

Categoric data are presented as number (%), continuous data are presented as mean with SD or median with IQR, depending on variable distribution

Table 2 Influence of use of azole antifungals on Tac metabolism

	Week 1 (n = 81)		Week 2 (n = 84)		Week 3 (n = 86)		Week 4 (n = 81)	
	Antifungals (-)	(+)	Antifungals (-)	(+)	Antifungals (-)	(+)	Antifungals (-)	(+)
n	70	11	67	17	63	23	55	26
Concentration (ng/mL)	5.8 (2.4– 10.2)	11.0 (9.3– 17.4) *	6.6 (5.3– 8.9)	10.4 (7.1– 13.6) *	7.4 (6.2– 9.9)	8.4 (6.9– 10.9)	7.7 (6.3– 9.0)	8.8 (6.4– 10.5)
Dose (mg/d)	2.0 (2.0– 3.0)	1.0 (1.0– 1.5) *	3.5 (2.0– 5.5)	0.5 (0.5– 2.0) *	4.0 (2.5– 6.0)	1.0 (0.5– 1.0) *	4.5 (2.5– 6.5)	1.0 (0.5– 1.0) *
C/D (ng/mL per mg/d)	134.10 (68.42– 238.99)	693.00 (417.60– 1000.50) *	129.00 (71.84– 230.14)	745.20 (506.35– 1148.35) *	122.01 (62.66– 215.78)	678.00 (320.17– 938.40) *	121.60 (59.40– 187.25)	525.60 (277.48– 885.96) *

Categoric data are presented as number (%), continuous data presented as mean with SD or median with IQR, depending on variable distribution. * indicates the level of significance ($p < 0.05$)

Table 3 Odds ratio (OR) with 95% confidence interval (CI) for risk factors of AKI incidence

Variables	Univariate analysis		Multivariate analysis	
	OR (95% CI)	<i>p</i>	OR (95% CI)	<i>p</i>
Age	0.96 (0.91–1.02)	0.179		
SCr, $\mu\text{mol/L}$	0.98 (0.95–1.01)	0.192		
Uric acid, $\mu\text{mol/L}$	1.00 (1.00–1.01)	0.088		
RBC, $\times 10^{12}$ cells/L	1.71 (0.80–3.66)	0.165		
Week 1 Tac concentration ≥ 7.5 ng/ml	5.38 (1.40–20.69)	0.014	5.38 (1.40–20.69)	0.014

Figures

Figure 1

(a) Tac concentration trend-lines of LTx patients during POD 3–28 (n = 90, 685 trough concentrations). Green shaded frame indicates the target range 8–10 ng/mL. Influence of CYP3A5 genotype on Tac concentration (b), dose (c) and C/D (d). Boxes and black lines in boxes represent IQR and median values, respectively; whiskers represent highest and lowest values. * indicates $p < 0.05$

Figure 2

Influence of CYP3A5 genotype and use of azole antifungals on Tac concentration in post-operative week 1 (a) (n = 81, CYP3A5*1 carriers: n = 37, *3/*3: n = 44), week 2 (b) (n = 84, CYP3A5*1 carriers: n = 40, *3/*3: n = 44), week 3 (c) (n = 86, CYP3A5*1 carriers: n = 39, *3/*3: n = 47) and week 4 (d) (n = 81, CYP3A5*1 carriers: n = 37, *3/*3: n = 44). CYP3A5*1 carriers had higher concentration in week 1 compared with CYP3A5*3/*3 group (Mann–Whitney U test). Use of azole antifungals did not change the influence of CYP3A5 genotype on Tac concentration. * indicates p < 0.05

Figure 3

Kaplan-Meier survival curves stratified by CYP3A5 genotypes at 30-day (a) and 1-year (b)

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- [Supplementaryfiles.docx](#)