Conversion of Twice-Daily Tacrolimus to Once-Daily Tacrolimus Formulation in Stable Pediatric Kidney Transplant Recipients: Pharmacokinetics and Efficacy

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The pharmacokinetics, efficacy and safety of once-daily tacrolimus formulation (Tac-OD) were assessed in 34 stable pediatric kidney transplant recipients. Enrolled patients received their dose of twice-daily tacrolimus formulation (Tac-BID) on study Days 0 through 7. On the morning of study Day 8, the total daily doses for patients were converted to Tac-OD on a 1:1 basis and maintained on a once-daily morning dosing regimen. Tacrolimus pharmacokinetic profiles were obtained on study Days 7, 14 and 28 (after dose adjustment). Although the mean C0 concentrations (4.10 ± 1.16–3.53 ± 1.10 ng/mL, p = 0.004), and AUC0–24 (151.8 ± 41.6–129.8 ± 39.3 ng h/mL, p < 0.001) were decreased significantly after a 1:1 based conversion, there was high interindividual variability. The dose of Tac-OD was decreased in 26.5% and increased in 44.1% of patients. The resultant tacrolimus dose and pharmacokinetic profiles on study Day 28 were comparable to those on Day 7. There were no serious adverse events. In conclusion, Tac-BID can be safely converted to Tac-OD in stable pediatric kidney transplant patients with the heightened therapeutic drug monitoring. Effects of drug conversion on the cardiovascular risk factors, neurological side effects and adherence should be further evaluated.

Key words: Conversion, pediatric kidney transplantation, pharmacokinetics, tacrolimus

Introduction

Nonadherence to prescribed immunosuppressive medication regimens is recognized as a leading cause of preventable graft loss for both adult and pediatric kidney transplant recipients (1,2). In the literature, up to 55% of renal transplant recipients have been reported to be nonadherent to immunosuppressive regimes, with adolescent and pediatric patients being more at risk (3,4). Therefore, effective interventions that improve patient adherence to immunosuppressive medications could be a core strategy to improve graft outcome after kidney transplantation in pediatric as well as adult patients (5,6).

Because a significant relationship of higher dosing frequencies to decreased adherence has been demonstrated (3), once daily morning administration of twice-daily tacrolimus formulation (Prograf®; Astellas Pharma, Tokyo, Japan; hereafter referred as Tac-BID) has been tried (7). Recently, a modified-release once-daily tacrolimus formulation (Advagraf®; Astellas Pharma, Tokyo, Japan; hereafter referred as Tac-OD) has been developed to provide more convenient once daily dosing that may contribute to improve patient adherence to immunosuppressive medication and by inference, graft survival, compared with twice-daily tacrolimus formulation. Robust data have been obtained on the pharmacokinetics of Tac-OD in adult transplant recipients. Most studies have shown consistent results of reductions in Cmin in stable and de novo patients with Tac-OD (8–12). Although an issue of adherence is most important in pediatric or adolescent transplant recipients, only few researches have been published on Tac-OD use in these age groups (13–15). In contrast to adult recipients, the difference in tacrolimus exposure following conversion from Tac-BID to Tac-OD was not apparent in industry-designed phase II pharmacokinetic trial in pediatric liver transplant patients (13).
As more data are required for the routine use of Tac-OD in pediatric transplant recipients, this present study was performed to clarify: (i) changes in pharmacokinetic parameters following conversion from Tac-BID to Tac-OD, (ii) requirement of tacrolimus dose change after conversion and (iii) clinical efficacy and safety of Tac-OD in stable pediatric kidney transplant recipients.

Materials and Methods

Patients
Pediatric ABO-compatible kidney transplant recipients (5–15 years) were eligible for enrollment if they had received kidney transplantation at least 1 year prior to enrollment and were receiving Tac-BID-based treatment. Recipients were required to have a stable renal function within at least 2 weeks prior to enrollment with a calculated glomerular filtration rate (GFR) > 40 mL/min/m² using the Schwartz formula (16) on a screening visit and to show tacrolimus whole blood trough levels between 3 and 20 ng/mL measured on at least two separate occasions and at least 1 week apart within 30 days prior to enrollment. Exclusion criteria included the following: experience of acute rejection episode within 90 days before the study entry, experience of acute rejection episode requiring antilymphocyte antibody therapy in the last 6 months, more than two rejection episodes within 12 months prior to study enrollment, aspartate aminotransferase or alanine aminotransferase higher than two times the upper limit of normal range, gastrointestinal disorder at the time of the pharmacokinetic study, a recipient of ABO-incompatible or cross match positive kidney transplantation, a recipient of multiorgan transplantation and current use of sirolimus.

The study (ClinicalTrials.gov NCT01476488) was conducted in accordance with the Declaration of Helsinki, Good Clinical Practice and the International Conference on Harmonisation guidelines and Declaration of Istanbul. The protocol was approved by the Seoul National University Hospital Institutional Review Board (H-1010-056-336). Patients gave written informed consent before study enrollment.

Study design
This phase III, open label, single-center, prospective study was conducted at the Seoul National University Hospital. All enrolled patients received their current dosage of tacrolimus from study Day 1 and the evening of Day 7. After completion of the first 24 h pharmacokinetic study on Day 7, patients were converted to Tac-OD on a mg:mg basis for their total daily dose on the morning of Day 8. The second pharmacokinetic study for Tac-OD was carried out on Day 14. On Days 15 and 21, the doses of Tac-OD were adjusted to achieve pre–switching tacrolimus C₀ levels measured on Day 7. The third pharmacokinetic study was conducted on Day 28. The entire study has 6-month extension period for evaluation of safety and effectiveness (Figure 1).

Pharmacokinetic analysis
For pharmacokinetic study, enrolled patients were admitted to Clinical Trials Center on the morning of Days 7, 14 and 28. Serial blood samples were collected for pharmacokinetic profiles at 0 (predose), 0.5, 1, 2, 4, 6, 8, 12, 12.5, 13, 14, 16, 18, 20 and 24 h. The trough level as the lowest concentration just before tacrolimus administration (C₀), the peak tacrolimus concentration (Cₘₐₓ) and the time required to reach Cₘₐₓ (Tₘₐₓ) for each subject were obtained directly from the raw data. The AUC₀–2₄₄ was calculated using linear trapezoidal rules from 0 to 24 h.

Enrolled patients were asked to fast for at least 2 h before and 1 h after receiving tacrolimus and also to take tacrolimus at a fixed time. On the day of the pharmacokinetic study, patients took tacrolimus 1 h before or 2 h after meals and the same food was served for lunch and dinner. Enrolled patients were instructed to avoid grapefruit and grapefruit juice during the study period.

Tacrolimus concentrations were determined, using high-performance liquid chromatography tandem mass spectroscopy using a Waters 2795 Alliance HT system (Micromass, Manchester, UK). The intraday coefficient of variation (CV) ranged from 5.2% to 9.3% and the accuracy was 96.0–104.0%. The interday CV varied from 3.6% to 9.6%. The lower limit of quantitation for tacrolimus was 0.8 ng/mL.

Figure 1: Study design and schedule of pharmacokinetic profiles (ClinicalTrials.gov NCT01476488). Tac-BID, twice-daily tacrolimus; Tac-OD, once-daily tacrolimus.
Once-Daily Tacrolimus in Pediatric Patients

Results

Patients

A total of 38 patients were enrolled in the study. Four patients were excluded from the pharmacokinetic evaluation because of protocol violation or incomplete pharmacokinetic profiles resulting from poor venous access. All 34 patients had evaluable pharmacokinetic profiles on all of the three time points as well as the safety data for the 6 months postconversion.

The mean age was 12.3 ± 2.8 years (range: 7.0–15.9 years) and the patients in this study received their grafts 40.3 ± 22.3 months before conversion to Tac-OD (Table 1). None of the patients had hyperlipidemia or diabetes mellitus, and 18 patients (52.9%) had received anti-hypertensive medications at baseline. No patients had neurological symptoms such as insomnia or tremor at study entry. None of the patients received non-immunosuppressive drugs that interact with tacrolimus.

The total daily dose of Tac-BID was 3.69 ± 1.40 mg (0.10 ± 0.04 mg/kg) before drug conversion. Thirty-three (97.1%) patients received mycophenolate mofetil, one patient (2.9%) was administered azathioprine and seven patients (20.6%) received prednisone at the time of study entry. All of the patients receiving adjunctive immunosuppressive medications continued to receive the same doses of their medications throughout the study period.

Table 1: Summary of patient baseline characteristics

<table>
<thead>
<tr>
<th>Patient characteristics</th>
<th>n = 34</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male, n (%)</td>
<td>20 (58.8)</td>
</tr>
<tr>
<td>Living donation, n (%)</td>
<td>18 (52.9)</td>
</tr>
<tr>
<td>Height (cm), mean (range)</td>
<td>143.7 (107.7–170.0)</td>
</tr>
<tr>
<td>Weight (kg), mean ± SD</td>
<td>40.7 ± 15.3</td>
</tr>
<tr>
<td>BMI, mean ± SD</td>
<td>19.0 ± 3.8</td>
</tr>
<tr>
<td>Age at conversion (years)</td>
<td>12.3 ± 2.8</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>12.5</td>
</tr>
<tr>
<td>Median</td>
<td>7.0–15.9</td>
</tr>
<tr>
<td>Time from transplantation to conversion (months)</td>
<td>40.3 ± 22.3</td>
</tr>
<tr>
<td>Range</td>
<td>34.0</td>
</tr>
<tr>
<td>Pre-existing conditions (%)</td>
<td>14.0–106.0</td>
</tr>
<tr>
<td>Diabetes/hypertension/hyperlipidemia</td>
<td>0/52.9/0</td>
</tr>
<tr>
<td>Total daily dose of prograf at baseline (mg)</td>
<td>3.69 ± 1.40</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>4.0</td>
</tr>
<tr>
<td>Median</td>
<td>1.5–6.0</td>
</tr>
</tbody>
</table>

Adjunctive immunosuppressive regimen

Mycophenolate mofetil, N (%) | 33 (97.1) |
Prednisolone, N (%)           | 7 (20.6) |
Azathioprine, N (%)           | 1 (2.9) |
respectively, after drug conversion on a 1:1 basis for total daily dose. The peak blood concentration of tacrolimus was also significantly decreased to 11.4 ± 4.0 ng/mL (p = 0.001), as expected. However, changes in pharmaco-
kinetic parameters were highly variable in the individual cases (Figure S1). Tacrolimus trough concentration and even Cmax was increased in nine patients. Therefore, in more than 70% of patients, the Tac-OD dose needed to be adjusted to achieve preconversion tacrolimus trough concentration and only 10 (29.4%) of the 34 pediatric patients did not require a dose adjustment; the dose was increased for 15 (44.1%) patients (3.3 ± 1.4–3.9 ± 1.4 mg/day, p < 0.001) and was decreased for 9 (26.5%) patients (4.7 ± 1.4–3.7 ± 1.5 mg/day, p = 0.021). After dose adjustment, C0 (3.87 ± 1.06 ng/mL) and AUC0–24 (144.9 ± 37.1 ng h/mL) on Day 28 were comparable with those on Day 7. Interestingly, the dose of Tac-OD (3.66 ± 1.32 mg/day) was similar with that of preconver-
sion Tac-BID (3.69 ± 1.40 mg/day, p = 0.843) despite dose adjustment in more than 70% of patients (Figure 2).

Patients with CYP3A5*3/*3 genotype (CYP3A5 nonex-
pressers) tended to experience significantly decreased dose-normalized C0 and AUC0–24 after mg:mg-based drug conversion (Table 2). No other clinical characteristics including age, body weight and body mass index were associated with tacrolimus dose change.

**Tacrolimus pharmacokinetics**

There was a similar correlation between AUC0–24 and Cmin for both tacrolimus formulations, with the correlation coefficient for Tac-BID (0.784) on study Day 7 being similar to Tac-OD (0.754) on Day 14 (Figure S2).

The equivalence of tacrolimus exposure was not demon-
strated based on lnAUC0–24 at steady state (Table 3); the ratio of lnAUC0–24 for Tac-OD/Tac-BID was 84.7 with 90% CI of 79.1–90.8% after mg:mg conversion. In spite of dose adjustment, the ratio of lnAUC0–24 was 87.8 (90% CI: 78.1–98.9%) on study Day 28. These ranges of CIs did not meet the accepted limits of 80–125% for equivalence. lnCmin and lnCmax were also not demonstrated to be equivalent; the 90% CIs were 78.7–93.0% and 70.7–89.1%, respectively, on Day 14.

The mean dose-normalized IV of the Tac-OD oral clearance was 21.4 ± 7.6%, and this was significantly higher than that of the Tac-BID oral clearance (16.5 ± 10.2%, p = 0.033).

**Clinical results**

There were no cases of discontinuation of Tac-OD, acute rejection, clinically indicated allograft biopsy, graft loss or

<table>
<thead>
<tr>
<th>CYP3A5 expresser (n = 10)</th>
<th>CYP3A5 nonexpresser (n = 17)</th>
</tr>
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<tbody>
<tr>
<td></td>
<td>Day 7</td>
</tr>
<tr>
<td>Dose/weight (mg/kg)</td>
<td>0.121 ± 0.045</td>
</tr>
<tr>
<td>C0 (ng/mL)</td>
<td>3.87 ± 1.06</td>
</tr>
<tr>
<td>Cmax (ng/mL)</td>
<td>13.7 ± 5.4</td>
</tr>
<tr>
<td>Tmax (h)</td>
<td>1.1 ± 0.3</td>
</tr>
<tr>
<td>AUC0–24 (ng h/mL)</td>
<td>149.3 ± 36.6</td>
</tr>
<tr>
<td>Dose-normalized C0 (ng/mL/mg/kg)</td>
<td>38.1 ± 19.9</td>
</tr>
<tr>
<td>Dose-normalized Cmax (ng/mL/mg/kg)</td>
<td>137.5 ± 88.5</td>
</tr>
<tr>
<td>Dose-normalized AUC0–24 (ng h/mL/mg/kg)</td>
<td>1460.5 ± 769.6</td>
</tr>
</tbody>
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Figure 2: Summary of mean blood tacrolimus concentra-
tion-time profiles and tacrolimus pharmacokinetic parameters for Day 7 (Tac-BID), Day 14 (Tac-OD, 1 week after mg:mg conversion) and Day 28 (Tac-OD, after Tac-OD dose-
adjustment). Tac-BID, twice-daily tacrolimus; Tac-OD, once-
daily tacrolimus. *p < 0.05, **p < 0.01; compared with Day 7.
Mean eGFR was 79.6 ± 27.0 mL/min at the end of the study and this was comparable to the baseline renal function (77.8 ± 27.9 mL/min, p = 0.223). Drug conversion to Tac-OD had a positive effect on hypertension. The amount of anti-hypertension medication administered was significantly decreased from 0.65 ± 0.8 to 0.5 ± 0.7 (p = 0.007), with blood pressure remaining stable throughout the 6-month postconversion period. The fasting serum glucose level was slightly decreased from 93.6 ± 7.8 to 90.7 ± 7.9 mg/dL (p = 0.061) in spite of no cases of diabetes mellitus.

No AEs were reported and no patients needed to discontinue Tac-OD due to AEs during the 6-month postconversion period. There were also no remarkable findings in clinical laboratory profiles including blood urea nitrogen, serum albumin, total bilirubin, alkaline phosphatase, alanine aminotransferase, aspartate aminotransferase, hemoglobin, hematocrit, total cholesterol and low-density lipoprotein cholesterol.

### Adherence

All of 34 patients completed the modified ITBS. Drug conversion to Tac-OD had a beneficial effect on patients’ barrier to immunosuppressants adherence (Table S1). More than 70% of patients ‘strongly agree or agree’ with the statement ‘the immunosuppressant medication(s) affects my daily life’ before drug conversion. However, this answer changed to ‘disagree or strongly disagree’ in 63.6% of patients after drug conversion (p < 0.001). Tac-OD had a positive effect on the perceptual complexity of dosing frequency (p = 0.001), and on the burden of capsules (or tablets) number (p = 0.001).

### Discussion

In the previous pediatric liver study, Heffron et al. (13) showed the difference in exposure between Tac-BID and Tac-OD was not apparent while studies for adult transplant patients consistently showed that AUC0–24 and Cmin were lower for Tac-OD compared with Tac-BID (9,10). Therefore, some authors have suggested a possible modifying influence of young age on between-formulation pharmacokinetic variability (11). In this study, however, we clearly show for the first time that exceedingly high percentages of stable pediatric transplant patients experience highly variable changes in pharmacokinetic parameters when their medications are converted from Tac-BID to Tac-OD. Therefore, in more than 70% of patients, the tacrolimus dose needed to be changed to adjust the baseline tacrolimus exposure. The equivalence of tacrolimus exposure was not demonstrated at Day 14 (1 week after mg:mg conversion) and even at Day 28 (after Tac-OD dose adjustment). These findings in pediatric transplant patients are similar to those recently published studies in adult transplant patients (9,10).

The oral clearance of Tac-OD has been reported to be lower in patients with CYP3A5*3/*3 (15). In this study, the CYP3A5 genetic polymorphism is the only covariate that influences the pharmacokinetics of Tac-OD.
Conversion from Tac-BID to Tac-OD did not affect the mean $C_0$ and AUC$_{0-24}$ in patients carrying CYP3A5*1 allele but patients with CYP3A5*3/*3 had a significantly lower $C_0$, $C_{\text{max}}$ and AUC$_{0-24}$ after conversion (Table 2). This result is compatible with the result from the previous study in adult patients (21) and is partly explained by the evidence that suggests that the CYP3A5 messenger RNA and protein levels may be higher in the proximal small intestine (22). However, two studies with small number of patients have showed the opposite impact of CYP3A5 polymorphisms on the pharmacokinetics of Tac-OD, and tacrolimus exposure significantly decreased in CYP3A5 expressers in stable or de novo renal transplant patients (23,24). Further research into this controversial impact of CYP3A5 polymorphisms during formulation switching is required.

A high intraindividual pharmacokinetic variability of tacrolimus has been associated with a poor long-term transplant outcome (17,25). This study shows that IIV of dose-normalized $C_{\text{min}}$ in pediatric patients is significantly higher for Tac-OD compared with Tac-BID ($21.4 \pm 7.6\%$ vs. $16.5 \pm 10.2\%$, $p = 0.033$). Although this may be partially caused by the dose change in 70% of patients after drug conversion, transplant physicians need to pay particular attention to the within-subject variability of tacrolimus oral clearance during Tac-OD therapy in pediatric transplant patients.

Despite the variable changes in tacrolimus pharmacokinetic parameters and significant dose adjustment when converting to Tac-OD, no serious AEs were shown. In addition, some benefits were shown in cardiovascular risk factors. Impaired insulin secretion from pancreatic beta cells, changes in hemodynamics and intense renal vasoconstriction can be caused by tacrolimus. Given these effects are related to the tacrolimus dose and concentration, Tac-OD may have beneficial effects on glucose metabolism and blood pressure due to low peak concentration compared with Tac-BID (26–28). In the clinical perspectives, these positive effects are quite important because cardiovascular disease is a leading cause of death after kidney transplantation in pediatrics as well as adults (29,30). Considering that Tac-OD can improve patient adherence to immunosuppressants by reducing barriers to adherence (Table S1), the beneficial effects of Tac-OD in the long-term outcome is certainly worth investigating.

This study has several limitations such as the small study population, the single ethnic group and short follow-up period. Because no patient had neurological side effects at baseline, effect of drug conversion on those side effects of tacrolimus was not able to be investigated in this study and this should be evaluated in future studies. The survey data should be interpreted with caution because the modification is not validated and it is not clear how many patients take medication on their own. However, this study clearly shows that the conversion of Tac-BID to Tac-OD results in highly variable changes in pharmacokinetic parameters of tacrolimus in stable pediatric kidney transplant patients.

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## Disclosure

The authors of this manuscript have no conflicts of interest to disclose as described by the American Journal of Transplantation.

## References


