Abbreviated Tacrolimus Area-Under-the-Curve Monitoring for Renal Transplant Recipients

Kim Ming Wong, FACP, Chi Chung Shek, FRCPA, Ka Foon Chau, FRCP, and Chun Sang Li, FRCP

● **The area under the concentration time curve (AUC) for oral tacrolimus (FK) may provide a more precise model for FK monitoring after renal transplantation. The purpose of this study is to identify a simple, cost-effective method for predicting FK AUC. FK concentrations were measured at 0, 1, 2, 4, 6, 8, and 12 hours after the morning dose. The predicted AUCs (AUC_ps) derived from regression equations were used to estimate the actual 12-hour AUCs (AUC_{12}s). The relationship between AUC_p and AUC_{12} was validated by determining the coefficient of multiple determination (R^2), percentage of prediction error (PE%), and percentage of absolute prediction error (APE%). Eighteen stable Oriental renal transplant recipients (9 men, 9 women) with a mean age of 42.6 ± 6 years and mean body weight of 62.7 ± 10 kg were recruited for the study. The FK AUC_{12}, trough, 2-hour, and 4-hour concentrations were 125 ± 24 h · ng/mL (range, 87.7 to 181.9 h · ng/mL), 6 ± 1.3 ng/mL, 18.1 ± 4.7 ng/mL, and 11 ± 2.4 ng/mL, respectively. Trough FK concentration did not have a significant correlation with AUC_{12} ($r = 0.34$; $P = 0.17$). AUC_p obtained by a two–time point regression equation using 2-hour (C2) and 4-hour (C4) FK concentrations: (AUC_P = 16.2 + 2.4*C2 + 5.9*C4) obtained an R^2, PE%, and APE% of 0.93, –0.2% ± 5.2% (range, –13% to 9.3%), and 3.6% ± 3.7% (range, 0.02% to 13%), respectively. We conclude that a two-point sampling method using C2 and C4 may be a more cost-effective FK monitoring strategy than morning FK trough levels in transplant recipients.**

INDEX WORDS: Area under the concentration time curve (AUC); tacrolimus (FK); pharmacokinetics; transplant.

TACROLIMUS (FK; Prograf; Fujisawa, Deerfield, IL) is a novel macrolide lactone with potent immunosuppressive properties. It has been shown to be effective as rescue or primary immunosuppressive therapy after organ transplantation.

Therapeutic monitoring of FK is recommended in view of the narrow therapeutic window to tailor an optimal dosage regimen for each patient. FK trough has been suggested to have a good correlation with the FK area under the concentration time curve (AUC) in renal transplant recipients during the immediate postoperative period.[1,2] However, it has been observed that the pharmacokinetics of FK change over time after transplantation.[3] The usefulness of FK trough levels in differentiating graft rejection episodes from nephrotoxicity has been questioned.[4,5] The same range of FK concentrations can be found in patients with graft rejection, toxicity, and stable graft function.[4,5]

The total drug effect on the body is better defined in terms of AUC.[6] AUC provides an individualized drug pharmacokinetic profile that reflects the total exposure of the drug in the body.[6-9] Traditionally, AUC is determined from six or more concentration-time data points. Its routine clinical use is limited by the need for multiple blood samples and drug measurements, which causes inconvenience to both patients and medical personnel. It has been suggested that abbreviated regression analysis using two or three time points after dosing can reliably predict the actual AUC in patients administered cyclosporine (CsA).[9] FK pharmacokinetic studies of renal transplant recipients who have a stable renal function are scarce in the literature.

This study aims to evaluate the pharmacokinetic profile of FK in an Oriental population and determine a practical abbreviated AUC monitoring strategy for FK for routine clinical use.

METHODS

Eighteen Oriental cadaveric renal transplant recipients (9 women, 9 men) with a mean age of 42.6 ± 6.0 (SD) years who were administered FK for primary prophylactic or rescue therapy for at least 6 months were studied after informed consent was obtained. None of the patients was receiving medication that would alter the metabolism of FK by inhibiting or inducing the cytochrome P-450 3A4 system. Patients were also advised to avoid grapefruit juice after transplantation, which might affect FK level. The standard immunosuppressive regimen in our center was composed of oral FK, adjusted to keep the trough FK level at 4 to 8 ng/mL; prednisolone, 5 to 7.5 mg/d; and azathioprine, 1.5 mg/kg/d. Pharmacokinetic profiles were obtained when the

From the Renal Unit, Department of Medicine, and the Department of Pathology, Queen Elizabeth Hospital, Hong Kong.

Received August 16, 1999; accepted in revised form October 29, 1999.

Address reprint requests to Kim Ming Wong, MD, Renal Unit, Department of Medicine, Queen Elizabeth Hospital, Kowloon, Hong Kong. E-mail: wongfkm@hotmail.com

0272-6386/00/3504-0012$3.00/0

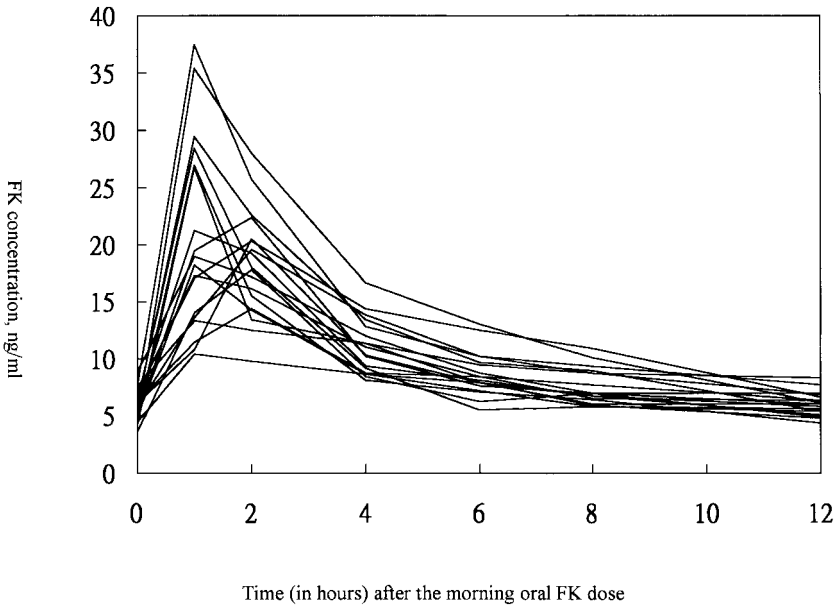

Fig 1. Relationship between FK concentration and time after FK administration.

patients were receiving stable doses of FK for at least 2 weeks before the study.

FK was administered orally in two equally divided doses at a fasting state in the morning (>12 hours from the last oral dose of FK) and 1 hour before meals to avoid other factors that might affect the bioavailability of FK. The 12-hour FK pharmacokinetic profile of each patient after the morning dose was analyzed. Ethylenediaminetetraacetic acid blood samples were collected at 0 (C0), 1 (C1), 2 (C2), 4 (C4), 6 (C6), 8 (C8), and 12 hours (C12) after the morning dose. Whole-blood FK level was measured by microparticle enzyme immunoassay using monoclonal antibody (IMx Tacrolimus II assay; Abbott Laboratories, Abbott Park, IL). The lower detection limit was 1.5 ng/mL, and the coefficients of variation were 15% at 5 ng/mL, 6.6% at 11 ng/mL, and 3.9% at 22 ng/mL.

Twelve-hour AUC (AUC_{12}) for each patient was calculated from a plot of FK concentrations versus time from 0 to 12 hours postdose using linear trapezoid rule. Abbreviated sampling equations were derived by multiple, stepwise regression analyses performed using AUC_{12} as the dependent variable and FK levels at different time points as the independent variables. The minimum number of time points sufficient to obtain a good estimate of the AUC_{12} was determined. The variance in the strength of association between predicted AUC (AUC_P) and AUC_{12} was reflected by the linear regression coefficient of multiple determination (R^2).

The AUC_Ps derived from the regression equations were validated by determining the predictive performance as described by Sheiner and Beal.[10] Prediction bias was measured by the percentage of prediction error (PE%; the degree of any given predictor tended to aim high or low relative to AUC_{12}), using the following equation:

$$PE\% = 100\% \times (AUC_P - AUC_{12})/AUC_{12}$$

Prediction precision was measured by the percentage of absolute prediction error (APE%; the magnitude of the difference between the AUC_P and AUC_{12}), using the following equation:

$$APE\% = 100\% \times |(AUC_P - AUC_{12})|/AUC_{12}$$

An error in prediction of approximately 15% was considered clinically acceptable because this was often chosen as the limit that would initiate a dosage adjustment.[11] Data are presented as mean ± SD. Statistical significance is defined as P less than 0.05. All statistical analyses were performed using SPSS software (version 7.5 for Windows 95; SPSS, Chicago, IL).

RESULTS

The median time after renal transplantation in the 18 patients was 2 years (range, 1 to 8 years).

Table 1. Correlation Between AUC_{12} Obtained From Full FK Pharmacokinetic Profile and FK Whole-Blood Concentrations at Different Time Points After Oral Dose

Time Points	Mean FK Concentration (ng/mL)	r	r^2	P
C0	6.0 ± 1.3 (3.6-8.9)	0.34	0.11	0.17
C1	20.6 ± 8.3 (10.4-37.5)	0.69	0.48	<0.001
C2	18.1 ± 4.7 (9.8-28)	0.85	0.72	<0.001
C4	11 ± 2.4 (8.1-16.7)	0.9	0.81	<0.001
C6	8.6 ± 2.0 (5.5-13)	0.82	0.67	<0.001
C8	7.5 ± 1.6 (5.8-10.9)	0.81	0.66	<0.001
C12	6.0 ± 1.0 (4.4-8.3)	0.49	0.24	0.04

NOTE. Mean AUC_{12} = 125 ± 24 h · ng/mL. Values expressed as mean ± SD (range).

Abbreviations: r, correlation coefficient; r^2, coefficient of determination.

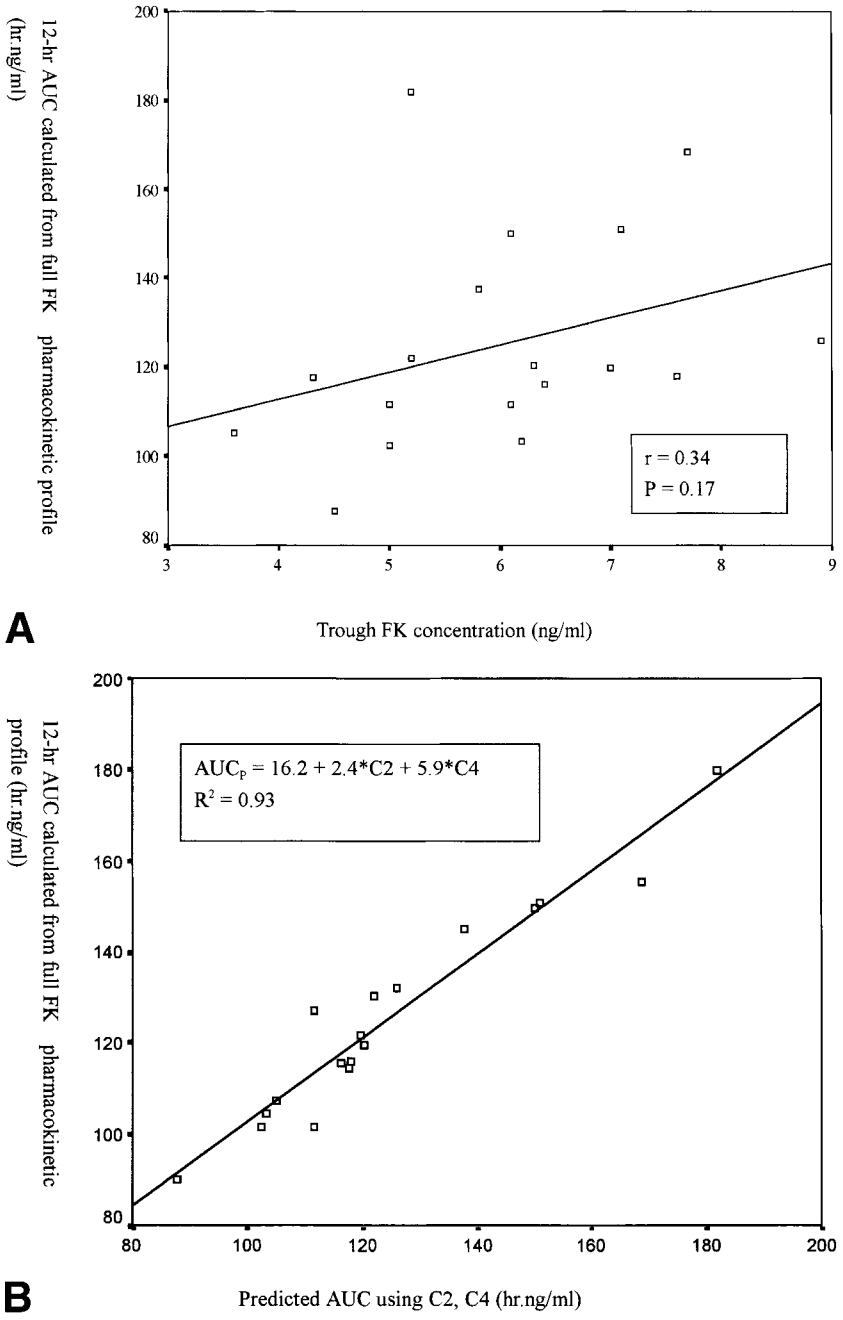

Fig 2. (A) Correlation between whole-blood trough FK concentration and actual 12-hour FK AUC. (B) Correlation between predicted FK AUC based on C2 and C4 and actual 12-hour FK AUC.

Mean body weight was 62.7 ± 10 kg, and mean FK dose was 0.09 ± 0.05 mg/kg. All patients had stable renal function, with a mean serum creatinine level of 1.42 ± 0.58 mg/dL, and no evidence of gastrointestinal dysfunction. The 12-hour pharmacokinetic profiles for FK in the 18 stable renal transplant recipients are plotted in Fig 1. The pharmacokinetics of FK were not affected by age or sex. As expected, interpatient variability of the pharmacokinetics of FK was high. The mean whole-blood FK trough (C0), 2-hour (C2), 4-hour (C4), and AUC_{12} levels were 6.0 ± 1.3 ng/mL, 18.1 ± 4.7 ng/mL, 11 ± 2.4 ng/mL, and 125 ± 24 hr · ng/mL (range, 87.7 to 181.9 hr · ng/mL), respectively. The relationship between different FK whole-blood concentrations at different time points and AUC_{12} is listed in Table 1. Our results showed that FK trough

Table 2. Regression Equations of the Relationship Between Predicted AUC and Actual 12-Hour AUC and the Associated Coefficient of Multiple Regression Values

Equations	Time Points	Regression Equations	R^2
A	C0, C1, C2, C4, C6, C8, C12	$AUC_P = 1.0 + 0.5*C0 + C1 + 1.5*C2 + 2*C4 + 2*C6 + 2.9*C8 + 2*C12$	1.0
B	C0, C1, C2, C4, C6, C8	$AUC_P = 5.4 + 1.1*C0 + C1 + 1.4*C2 + 2.3*C4 + 2*C6 + 3.3*C8$	0.99
C	C0, C1, C2, C4, C6	$AUC_P = 8.3 + 1.2*C0 + 0.9*C1 + 1.6*C2 + 2.7*C4 + 3.7*C6$	0.99
D	C0, C1, C2, C4	$AUC_P = 10 + 1.4*C0 + 0.8*C1 + 1.6*C2 + 5.5*C4$	0.98
E	C0, C2, C4	$AUC_P = 13.3 + 1.2*C0 + 2.4*C2 + 5.6*C4$	0.93
F	C2, C4	$AUC_P = 16.2 + 2.4*C2 + 5.9*C4$	0.93
G	C0, C1, C2	$AUC_P = 24.5 + 3.8*C0 + 0.9*C1 + 3.3*C2$	0.82
H	C1, C2	$AUC_P = 44 + 0.8*C1 + 3.5*C2$	0.77
I	C0, C2	$AUC_P = 26.2 + 3.6*C0 + 4.2*C2$	0.76
J	C0, C1	$AUC_P = 53.22 + 5.26*C0 + 1.95*C1$	0.57

level did not have a significant correlation with AUC_{12} ($r = 0.34$; $P = 0.169$; Fig 2A). The best correlation between a single time point and AUC_{12} was obtained at 4 hours (C4), which could explain 81% of the variance in AUC_{12} ($P < 0.001$).

The derived regression equations for predicting AUC_{12} and the associated R^2 values are listed in Table 2. There was a small-to-moderate degree of variance in the association between AUC_P and AUC_{12}, with R^2 ranging from 0.57 to 1. The minimal time points to obtain a reasonably good estimate of AUC_{12} ($R^2 = 0.93$) were two (equation F, using C2 and C4, Fig 2B).

Table 3 lists the PE% and APE% of each regression equation in predicting AUC_{12}. The PE% and APE% of equations A through J were within the 15% acceptable limit. The minimum number of time points to get an unbiased yet precise estimate of AUC_{12} were also two (equation F, using C2 and C4). The frequency distribution of PE% and APE% of equation F was plotted in histograms with 5% intervals. The PE% and APE% of equation F (using C2, C4) were –0.2% ± 5.2% (range, −13% to 9.3%; Fig 3A) and 3.6% ± 3.7% (range, 0.02% to 13%, Fig 3B), respectively.

DISCUSSION

Our results indicate that correlation between FK trough level and AUC_{12} is poor and may not be reliable in predicting the total drug exposure in renal transplant recipients. We have shown that a simple yet reasonably precise approach in estimating AUC_{12} using a two-point blood sampling strategy is feasible (equation F). As shown in the histograms (Fig 3A and B), PE% and APE% of equation F are within the 15% error limit and may provide an unbiased and precise model for the estimation of the total drug exposure.

Table 3. Data Showing the Mean PE% and APE% of the AUC_P From Regression Equations Compared With the AUC_{12} Obtained From the Full Pharmacokinetic Profile

Equations	PE%	APE%
A (C0, C1, C2, C4, C6, C8, C12)	−0.5 ± 0.3 (−1.6-−0.3)	0.5 ± 0.3 (0.3-1.6)
B (C0, C1, C2, C4, C6, C8)	−0.2 ± 1.1 (−2.2-1.6)	0.8 ± 0.7 (0.01-2.2)
C (C0, C1, C2, C4, C6)	0.5 ± 2.0 (−4.5-3.8)	1.7 ± 1.2 (0.2-4.5)
D (C0, C1, C2, C4)	−0.08 ± 3 (−3.7-4.9)	2.5 ± 1.6 (0.1-4.9)
E (C0, C2, C4)	−0.8 ± 5.1 (−8.9-14)	3.5 ± 3.8 (0.05-14)
F (C2, C4)	−0.2 ± 5.2 (−13-9)	3.6 ± 3.7 (0.02-13)
G (C0, C1, C2)	0.5 ± 8.1 (−11.8-14)	6.8 ± 4.2 (0.6-14)
H (C1, C2)	0.6 ± 9.3 (−16.1-13.8)*	7.6 ± 5.0 (0.8-16.1)*
I (C0, C2)	0.7 ± 9.5 (−16.3-21.0)*	7.7 ± 5.4 (0.06-21.0)*
J (C0, C1)	1.3 ± 12.0 (−19.9-22.4)*	9.5 ± 7.0 (1.0-22.4)*

NOTE. Values expressed as mean ± SD (range).
*Predicted AUC was outside the ±15% range from the actual AUC.

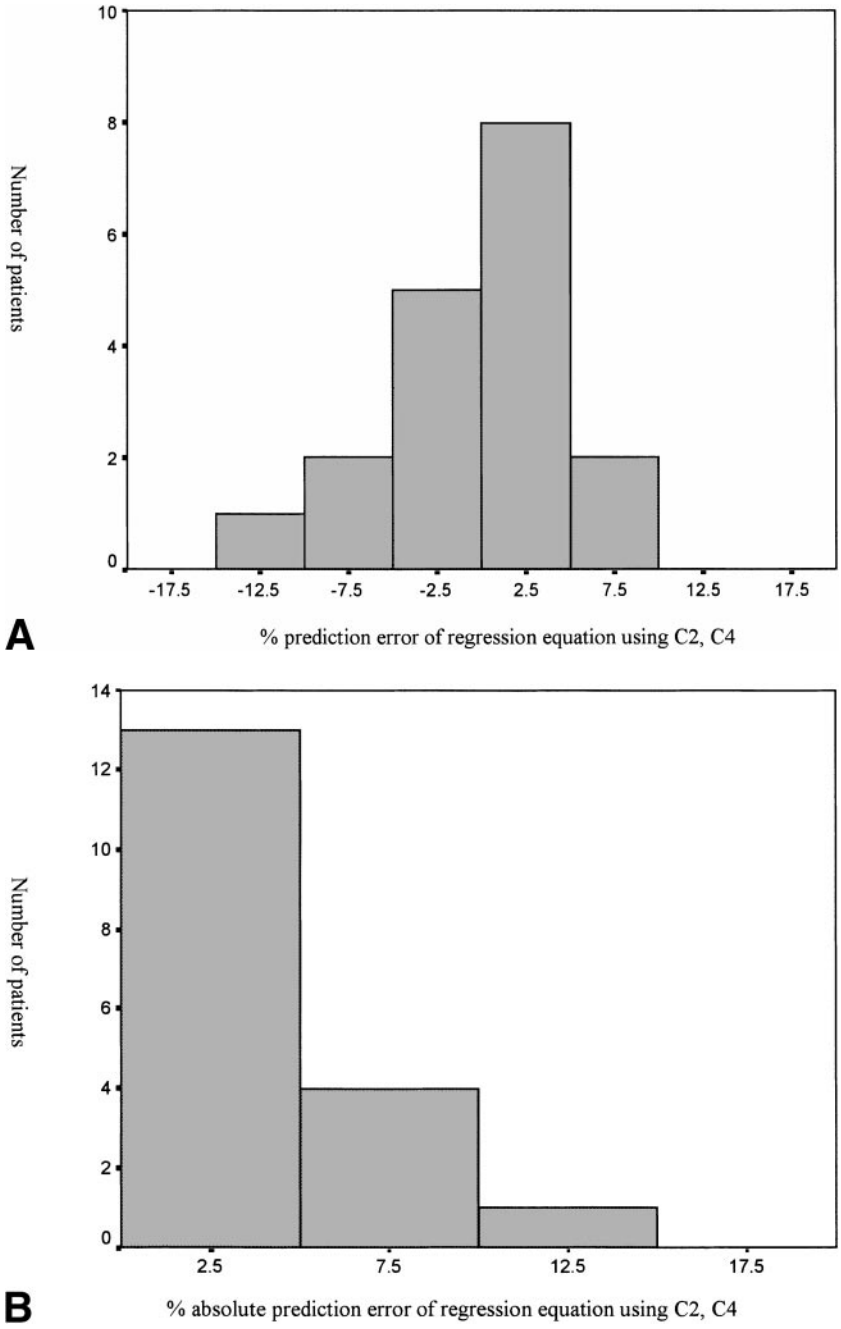

Fig 3. (A) Frequency distribution of the PE% of regression equation based on C2 and C4 and the actual 12-hour FK AUC. (B) Frequency distribution of the APE% of regression equation using C2 and C4 and the actual 12-hour FK AUC.

The unpredictability of the immunosuppressive drug exposure has been suggested to be related to graft rejection and suboptimal long-term renal allograft outcome.[12] Therapeutic drug monitoring has an important role in optimizing therapy. As with CsA, FK shows large interindividual variability in its bioavailability and has a relatively narrow therapeutic window.[13,14] Oral absorption is poor, with a bioavailability of approximately 25% and highly variable. The absorption is affected by multiple factors, including the presence of food, antacid, and altered gut metabolism.[15] The monitoring of drug levels can offer tremendous help to the clinicians.

Therapeutic monitoring of FK has traditionally been performed by adjustment of dosage to

achieve a certain target range of drug level. Morning trough concentration monitoring is by far the most commonly used tool to guide dosage adjustment.[16] Only few data concerning the pharmacokinetics of FK are available from renal transplant recipients with stable graft function.[1,2] A good correlation ($r = 0.92$) between AUC_{12} and the trough whole-blood level of FK has been observed in the early posttransplantation period.[1,2,17] Other investigators have shown that the use of morning tough-level monitoring in reflecting AUC_{12} has a r^2 of 0.35 only in liver transplant recipients.[16] Evening trough-level monitoring may provide more information ($r^2 = 0.71$) but may not be practical as a routine monitoring strategy because of the inconvenience to patients. The variation in bioavailability seen in our patients necessitates profile AUC monitoring because the trough and peak levels do not correlate well with AUC_{12} and thus are not true estimates of the actual drug exposure in the patients. Trough FK monitoring may therefore be suboptimal in predicting underdosage and drug toxicity.

An accurate and convenient monitoring strategy is deemed important for optimal immunosuppression. Clinical outcome was improved by AUC monitoring.[12,18] Kahan et al[19] proposed the use of an AUC method for monitoring CsA therapy that resulted in a decreased incidence of delayed graft loss during the first month after renal transplantation. Other investigators have reported an association between low AUC values and poor graft outcome.[20] It has been suggested that to avoid an unnecessarily high incidence of adverse events of FK and yet provide adequate immunosuppression, a dosing regimen with a target AUC_{12} of approximately 100 hr · ng/mL may be advisable.[21] However, the monitoring of full pharmacokinetic profiles by AUC has not gained much clinical acceptance in view of the associated cost to patients and medical personnel. Taking frequent blood samples over 12 hours is accurate but has obvious practical disadvantages. Despite the limitation of the use of trough level in reflecting total drug exposure, its use still predominates in most transplant centers.

We attempted to derive an abbreviated pharmacokinetic profile using a limited blood-sampling strategy to monitor total FK exposure in individual patients. To be clinically applicable, the estimated AUC based on a limited blood sampling strategy should have a good correlation with actual AUC and require a small number of blood samples with minimal waiting time for the patients. In our patients, a two–time point FK level sampling strategy ($AUC_P = 16.2 + 2.4*C2 + 5.9*C4$) is probably the most cost-effective method for FK monitoring. Correlation with AUC_{12}, PE%, and APE% is similar to that obtained from the regression equation using a three–time point sampling strategy. Patients are requested to stay in the clinic for only 2 hours for blood sampling, after taking the morning dose of FK at home.

In conclusion, we have shown that FK AUC_P predicted by a regression equation (using C2 and C4) can accurately estimate AUC_{12} calculated from the seven-point FK pharmacokinetic profile. It is more economical, and the inconvenience to patients is minimized. Results of FK AUC_{12} monitoring on long-term graft outcome are still lacking. Future studies are warranted to determine the optimal target range of FK AUC_{12} in renal transplant recipients.

REFERENCES

1. Mekki Q, Lee C, Aweeka F, Laskow D, Neylan J, Mendez R, Steinmuller D, Schechrer P: Pharmacokinetics of tacrolimus (FK506) in kidney transplant patients. Clin Pharmacol Ther 53:238A, 1993 (abstr)

2. Lee C, Jusko W, Shaefer M, Klintmalm G, Hebert M, Piergies A, Schechter P, Mekki Q: Pharmacokinetics of tacrolimus (FK506) in transplant patients. Clin Pharmacol Ther 53:181A, 1993 (abstr)

3. Christiaans M, van Duijnhoven E, Beysens T, Undre N, Schafer A, van Hooff J: Effect of breakfast on the oral bioavailability of tacrolimus and changes in pharmacokinetics at different times posttransplant in renal transplant recipients. Transplant Proc 30:1271-1273, 1998

4. Takahara S, Kakado Y, Kameoka H, Takano Y, Jiang H, Moutabarrik A, Ishibashi M, Okuyama A, Sonoda T: Monitoring of FK506 blood levels in kidney transplant recipients. Transplant Proc 26:2106-2108, 1994

5. Winkler M, Ringe B, Rodeck B, Melter M, Stoll K, Banmann J, Wonigeit K, Pichlmayr R: The use of plasma levels for FK506 dosing in liver graft patients. Transpl Int 7:329-333, 1994

6. Johnston A, Keown PA, Holt DW: Simple bioequivalence criteria: Are they relevant to critical dose drugs? Experience gained from cyclosporine. Ther Drug Monit 19:375-381, 1997

7. Grevel J, Welsh MS, Kahan BD: Cyclosporine monitoring in renal transplantation: Area under the curve monitoring is superior to trough level monitoring. Ther Drug Monit 11:246-248, 1989

8. Kahan BD, Welth M, Rutzky L: Challenges in cyclosporine therapy: The role of therapeutic monitoring by area

under the curve monitoring. Ther Drug Monit 17:621-624, 1995

9. Gaspari F, Perico N, Signorini O, Caraso R, Remuzzi G: Abbreviated kinetic profiles in area-under-the-curve monitoring of cyclosporine therapy. Kidney Int 54:2146-2150, 1998

10. Scheiner LB, Beal SC: Some suggestions for measuring predictive performance. J Pharmacokinet Biopharm 9:503-512, 1981

11. Meier-Kriesche HU, Kaplan B, Brannan P, Kahan BD, Portman RJ: A limited-sampling strategy for the estimation of eight-hour Neoral areas under the curve in renal transplantation. Ther Drug Monit 20:401-407, 1998

12. Lindholm AS, Kahan BD: Influence of cyclosporine pharmacokinetics, trough concentrations, and AUC monitoring on outcome after kidney transplantation. Clin Pharmacol Ther 54:205-218, 1993

13. Hooks MA: Tacrolimus, a new immunosuppressant, a review of the literature. Ann Pharmacother 28:501-511, 1994

14. Jusko WJ, Piekozewski W, Klintmalm GB, Shaefer MS, Hebert MI, Piergies AA, Lee CC, Schechter P, Mekki QA: Pharmacokinetics of tacrolimus in liver transplant patients. Clin Pharmacol Ther 57:281-290, 1995

15. Venkataramanan R, Swaminathan A, Prasad T, Jain A, Zuckerman S, Warty V, McMichael J, Lever J, Burckart G, Starzl T: Clinical pharmacokinetics of tacrolimus. Clin Pharmacokinet 29:404-430, 1995

16. Ku YM, Min DI: An abbreviated area-under-the-curve monitoring for tacrolimus in patients with liver transplants. Ther Drug Monit 20:219-223, 1998

17. Boswell GW, Bekersky I, Fay J, Wingard J, Autin J, Weisdorf D, Maher R, Fitzsimmons U, Nash R: Tacrolimus pharmacokinetics in bone marrow transplant patients. Bone Marrow Transplant 2:23-28, 1998

18. Bowles MJ, Water JB, Lechler RI, Williams G: Do cyclosporin profiles provide useful information in the management of renal transplant recipients? Nephrol Dial Transplant 11:1597-1602, 1996

19. Kahan BD, Welsh M, Rutzky L, Lewis R, Knight R, Katz S, Napoli K, Grevel J, Van Buren CT: The ability of pre-transplant test dose pharmacokinetic profiles to reduce each adverse event after renal transplantation. Transplantation 53:35-51, 1992

20. Schroeder TJ, Hariharan S, First MR: Variations in bioavailability of cyclosporine and relationship to clinical outcome in renal transplant subpopulations. Transplant Proc 27:837-839, 1995

21. Filler G, Grygas R, Mai I, Stolpe HJ, Greiner C, Bauer S, Ehrich JHH: Pharmacokinetics of tacrolimus (FK506) in children and adolescents with renal transplants. Nephrol Dial Transplant 12:1668-1671, 1997