A Predictive Model for Area Under the Concentration versus Time Curve of Cyclosporin A Using Several Routine Monitoring Results in Renal Transplant Patients

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We created a predictive model for the area under the concentration versus time curve (AUC) of cyclosporin A (CsA) using routine monitoring results, and examined its clinical utility. Based on 48 clinical time courses accumulated from renal transplant patients, the AUC predictive model was created. An estimate of the AUC
c0.08 (integrated from time zero to 8 h) was then given as follows: AUC
c0.08 = 5673.1 x log(TL) + 9342.8 x log(OB) + 64.1 x D
prd × 869.4 x DTK - 168.9 x HCT - 161.2 x SCG - 11.3 x GPT + 3.0 x PL - 588.6 x SEX - 24794.5. In this model, the AUC
c0.08 (ng·h/ml) is given as a function of the CsA trough levels (TL, ng/ml), obesity (OB, %), daily dose of prednisolone (D
prd, mg/d), donor type of kidney (DTK), hematocrit (HCT, %), serum creatinine (SCG, mg/dl), glutamate-pyruvate transaminase activity (GPT, IU/l), plasma lipids (PL, mg/dl) and sex distinction (SEX). The Statistical significance of this multiple regression was p < 0.00001 (R² = 0.862, n = 48), and the day after transplantation, neither the administered oral dose of CsA, or the patient’s age had any contribution to the regression. The predictive performance of this model was almost equal to that of the existing method which used 3-point data on the concentration versus time curve. In clinical adaptation for renal transplant patients, the steady-state concentration of CsA (C
ss) based on the AUC
c0.08 predictive model was significantly decreased during acute gastroenteritis or before acute rejection, whereas nephrotoxicity was increased, even though CsA trough levels were within a normal therapeutic range (100—200 ng/ml). These findings suggest that the created AUC
c0.08 predictive model using routine monitoring results, i.e., the trough level of CsA, biochemical tests, a daily dose of prednisolone (PRD), and basic patient information, is convenient as a monitoring device for CsA therapy, and is satisfactory in clinical practice.

Key words cyclosporin A; predictive model; AUC monitoring; trough level monitoring; disease state; renal transplantation

It is well known that the pharmacokinetics of cyclosporin A (CsA), a potent immunosuppressive agent, after organ transplantation exhibits very complex aspects.1,2 In addition to the type of transplanted organ, several physiological factors in relation to disease states after organ transplantation markedly affected the pharmacokinetics of oral CsA.3 A reasonable correlation has been noted between the concentration of CsA and the immunosuppressive response in various in vitro tests, but this correlation is much less obvious in vivo.4 Indeed, via trough level monitoring of CsA only, it is often difficult to distinguish between the clinical signs of its major adverse effects and the luck of therapeutic efficacy.5 Therefore, the area under the concentration versus time curve (AUC) monitoring of CsA had been recommended to adjust the oral CsA dose and to obtain the target concentrations of CsA throughout the monitoring periods.5-8 In addition, the concept of AUC monitoring for CsA therapy enables us to determine the steady-state concentration of CsA (C
ss) after oral administration, which provides a better measure of exposure to CsA than does a trough level of CsA. It is considered that presumption in the degree of exposure to CsA by the C
ss would lead to the monitoring of therapeutic efficacy or its tissue toxicity. However, the AUC monitoring requires multiple sample points after the oral administration of CsA, and the method is not suitable for routine monitoring because multiple sampling forces burden a patient.

For more precise use of the CsA trough level monitoring in clinical practice, we have continued to investigate the relationship between the pharmacokinetics of CsA and its distribution property in blood. In a series of previous reports,9-12 we found that the erythrocyte-to-plasma distribution ratio of CsA (CsA-EP) reflects an alteration in disease state and CsA pharmacokinetics. Therefore, the CsA-EP is a useful indicator to predict CsA pharmacokinetics under several disease states.12 For clinical convenience, we derived a predictive model of the CsA-EP using routine monitoring results, including the CsA trough level,10 and have used this as a monitoring device when the trough level monitoring is performed. As a continuation of these investigations, we demonstrated here an AUC predictive model using routine monitoring results, including CsA trough level, patients’ fundamental information, biochemical tests, and a daily dose of metabolic inducer, prednisolone, and then evaluated the clinical utility of this AUC predictive model.

MATERIALS AND METHODS

Patients and Variables for Multivariate Analysis to Create an AUC Predictive Model Forty-eight accumulated clinical time-courses of CsA, obtained from 13 renal transplant patients during the monitoring period from April 1990 to March 1996, were used for this analysis. The age range of analyzed patients was from 19 to 57 years (mean 37.2±13.1 years); 10 were male and 2 were female; 5 were cadaveric donors and 8 were living-related donor recipients. Immunosuppression after renal transplantation was achieved with triple therapy consisting of

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### Table 1. Description of Data Used in Creating an AUC Predictive Model in Renal Transplant Patients

<table>
<thead>
<tr>
<th>Patient No.</th>
<th>Age (year)</th>
<th>Donor/sex</th>
<th>DAT (d)</th>
<th>D&lt;sub&gt;ca&lt;/sub&gt; (mg/kg/d)</th>
<th>TL (mg/ml)</th>
<th>AUC&lt;sub&gt;0-t&lt;/sub&gt; (ng·h/ml)</th>
<th>GPT (IU/l)</th>
<th>SCR (mg/dl)</th>
<th>HCT (%)</th>
<th>PL (mg/dl)</th>
<th>D&lt;sub&gt;pred&lt;/sub&gt; (mg/d)</th>
<th>OR&lt;sup&gt;a&lt;/sup&gt; (%)</th>
<th>Clinical episodes&lt;sup&gt;b&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>7, 20, 48</td>
<td>L/ M</td>
<td>6.036 ± 1.567</td>
<td>116 ± 42</td>
<td>3920 ± 1397</td>
<td>18 ± 8</td>
<td>2.9 ± 0.2</td>
<td>32.4 ± 4.3</td>
<td>330 ± 64</td>
<td>28.1 ± 16.8</td>
<td>98.7 ± 4.8</td>
<td>A rej. (day 196)</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>7, 17, 30</td>
<td>L/ M</td>
<td>5.986 ± 1.093</td>
<td>145 ± 29</td>
<td>3044 ± 1297</td>
<td>45 ± 31</td>
<td>1.5 ± 0.2</td>
<td>31.6 ± 2.5</td>
<td>324 ± 54</td>
<td>30.5 ± 14.2</td>
<td>107.0 ± 2.8</td>
<td>HTx (day 9, 17)</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>7, 19, 29</td>
<td>C/ M</td>
<td>4.739 ± 1.410</td>
<td>113 ± 46</td>
<td>2110 ± 853</td>
<td>58 ± 49</td>
<td>6.9 ± 5.4</td>
<td>31.0 ± 1.5</td>
<td>319 ± 31</td>
<td>32.0 ± 12.7</td>
<td>109.7 ± 2.5</td>
<td>HTx (day 19, 29, 40)</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>12, 19, 29</td>
<td>C/ M</td>
<td>5.940 ± 0.193</td>
<td>325 ± 78</td>
<td>6884 ± 1176</td>
<td>30 ± 16</td>
<td>4.9 ± 3.4</td>
<td>23.1 ± 2.2</td>
<td>265 ± 72</td>
<td>30.0 ± 12.7</td>
<td>111.1 ± 3.1</td>
<td>HTx (day 29, 40)</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>36, 48, 56</td>
<td>L/ F</td>
<td>4.116 ± 0.792</td>
<td>347 ± 23</td>
<td>3953 ± 479</td>
<td>25 ± 19</td>
<td>1.2 ± 0.0</td>
<td>28.1 ± 1.1</td>
<td>300 ± 30</td>
<td>22.5 ± 2.5</td>
<td>79.0 ± 4.5</td>
<td>HTx (day 36)</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>7, 15, 26</td>
<td>L/ M</td>
<td>6.078 ± 0.378</td>
<td>148 ± 25</td>
<td>4209 ± 581</td>
<td>65 ± 59</td>
<td>1.0 ± 0.1</td>
<td>30.1 ± 1.3</td>
<td>314 ± 65</td>
<td>33.0 ± 12.0</td>
<td>95.6 ± 4.5</td>
<td>HTx (day 7, 15, 35)</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>19, 30, 40</td>
<td>C/ M</td>
<td>5.885 ± 0.128</td>
<td>164 ± 43</td>
<td>3863 ± 1045</td>
<td>13 ± 2</td>
<td>3.1 ± 3.5</td>
<td>28.3 ± 1.2</td>
<td>351 ± 18</td>
<td>23.0 ± 4.8</td>
<td>96.3 ± 2.1</td>
<td>None</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>7, 15, 24</td>
<td>L/ M</td>
<td>5.445 ± 1.815</td>
<td>254 ± 84</td>
<td>6190 ± 1764</td>
<td>30 ± 22</td>
<td>1.2 ± 0.1</td>
<td>35.5 ± 2.9</td>
<td>420 ± 77</td>
<td>30.0 ± 13.0</td>
<td>112.1 ± 3.8</td>
<td>NTx (day 24, 35, 55)</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>7, 14</td>
<td>C/ F</td>
<td>8.520 ± 358</td>
<td>388</td>
<td>6899</td>
<td>21</td>
<td>1.3</td>
<td>35.1</td>
<td>251.0</td>
<td>40.0</td>
<td>121.0</td>
<td>None</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>50</td>
<td>C/ F</td>
<td>5.000 ± 77</td>
<td>1615</td>
<td>40</td>
<td>11.6</td>
<td>23.0</td>
<td>304.0</td>
<td>45.0</td>
<td>106.2</td>
<td>None</td>
<td></td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>7, 44</td>
<td>L/ M</td>
<td>7.261 ± 1.029</td>
<td>103 ± 55</td>
<td>2763 ± 623</td>
<td>11 ± 10</td>
<td>1.9 ± 0.3</td>
<td>32.3 ± 2.4</td>
<td>338 ± 42</td>
<td>37.5 ± 13.9</td>
<td>86.2 ± 2.3</td>
<td>None</td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>160</td>
<td>C/ M</td>
<td>5.742 ± 137</td>
<td>1563</td>
<td>11</td>
<td>2.1</td>
<td>33.0</td>
<td>300.0</td>
<td>10.0</td>
<td>83.8</td>
<td>C rej. (day 160)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>13</td>
<td>26</td>
<td>L/ M</td>
<td>5.300 ± 266</td>
<td>4003</td>
<td>17</td>
<td>1.6</td>
<td>31.0</td>
<td>399.0</td>
<td>25.0</td>
<td>69.1</td>
<td>None</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

The numerical data with a sample size of more than 3 represent the mean ± S.D.  
<sup>a</sup> Number of time courses.  
<sup>b</sup> Day after transplantation when clinical time course study was performed.  
<sup>c</sup> The obesity was calculated by adding 100 to make a logarithmic expression.  
<sup>d</sup> Clinical episodes were cited from patients' medical records.  
<sup>e</sup> The numerical data with a sample size of 2 represent the mean.  
C, cadaveric donor; L, living-related donor; M, male; F, female; A rej., acute renal rejection; C rej., chronic renal rejection; HTx, hepatotoxicity; NTx, nephrotoxicity; ATN, acute tubular necrosis.

CsA, prednisolone (PRD), and azathiopurine or mizoribine according to a protocol of renal transplantation enforced at Shiga University of Medical Science. The daily oral dose of azathiopurine was maintained at 1.5 mg/kg/d throughout the monitoring periods. The daily oral dose of PRD was tapered from 50 mg/d to 10 mg/d at almost 10-d intervals. CsA (Sandimmun® soft gelatin capsule 25 mg or oral solution 100 mg/ml) was administered to patients in two divided doses at 12-h intervals. The daily oral dose of CsA was started from 8 mg/kg/d for living-related donor recipients, and for cadaveric donor recipients the dose was started from 6 mg/kg/d. Informed consent had been obtained from all the patients. Before renal transplantation, immunosuppressive therapy with CsA, less effect and adverse effects of CsA, and several clinical tests to evaluate clinical efficacy, including blood-collection for the CsA pharmacokinetic study, were explained to the patients by physicians. Individual data used for analysis are listed in Table 1. Blood samples were collected into heparinized tubes on or after the third day after a change in oral dose (steady-state). Sampling times for the accumulated 48 clinical time courses were uneven; therefore, we used the data from zero-time (tough level at 6:00 a.m.) to 8 h after the oral administration of CsA for the pharmacokinetic analysis. Biochemical tests were carried out at the same time as CsA trough level measurements in each time course. Renal function was assessed by serum creatinine level (SCR, mg/dl), and hepatic function by glutamate-pyruvate transaminase (GPT, IU/l). As CsA blood distribution-related factors, hematocrit (HCT, %) and plasma lipids (PL, mg/dl), which is equal to the sum of total cholesterol and triglycerides were used. The daily dose of CsA per body weight (D<sub>ca</sub>, mg/kg/d), the daily dose of PRD (D<sub>pred</sub>, mg/d), CsA trough levels (TL, ng/ml), age (AG, years), and day after transplantation (DAT, d) were used as CsA pharmakokinetic-related variables. Sex distinction (SEX), donor type of kidney (DTK), and obesity (OB, %) were used as physiological factors affecting CsA pharmacokinetics. The values for the SEX and DTK were given as binary data; namely, the male or cadaveric donors were equal to 1, and the female or living-related donors were equal to 2.

**Data Analysis and Statistics**  
The AUC from time zero to 8 h after the oral administration of CsA (AUC<sub>0→8</sub>, ng·h/ml) was calculated using a linear trapezoidal approximation. The OB for all patients was calculated using the equation as defined by OB = 100 × (BW/22BH<sup>2</sup> - 1) + 100, where BW and BH represent the body weight (kg) and body height (m), respectively, and 22BH<sup>2</sup> represents the standard body weight based on the body mass index. The CsA erythrocyte-to-plasma distribution ratio (CsA-EP) was given by the following equation: CsA-EP = 0.0831 - 0.2944 × PL - 0.0037 × TL - 0.0553 × HCT + 0.0845 × BW + 0.4447 × SCr - 0.0366 × AG. In this equation, the PL was given as a millimolar concentration. Multiple regression analysis, including bivariate regression analysis, was performed using the Seto/B software package (Kyouritsu Pub. Co., Ltd., Japan) on a PC-9801 Vmz microcomputer. The AUC<sub>0→8</sub> predictive model was created by a stepwise selection method, and the conformity of the model and the contribution of an independent variable for the model were checked by Akaike's information criteria (AIC). The regression
equation was considered significant if the p-value for the regression was less than or equal to 0.05 using the F-distribution test. Multicollinearity among independent variables in the multiple regression was checked by a variance inflation factor (VIF). The mean concentrations of CsA at steady-state (CSS, ng/ml) derived from our \( AUC_{0-8} \) predictive model was then calculated by dividing the predicted \( AUC_{0-8} \) by 8 h.

**Drug Assay** The CsA concentration in whole blood was measured by means of a monoclonal-fluorescence polarization immunoassay (m-FPIA) using the TDX operation system (Abbott Laboratory, North Chicago, IL, U.S.A.) according to the assay manual.\(^{15}\) The coefficient of variation of within-day and between-day precision were within 3.2% and 3.9%, respectively. The limit of detection was 25 ng/ml. Some of the CsA concentration versus time curves obtained in the early period were measured by high-performance liquid chromatography (HPLC).\(^{16}\) According to our research data inside the institution, a regression coefficient given by bivariate analysis of data between our HPLC and the m-FPIA was 0.93 (\( p < 0.001, n = 38 \)). The measurements by our HPLC, therefore, had one-to-one correspondence with that by the m-FPIA.

**Comparative Study** By comparing the CSS derived from our \( AUC \) predictive model with that from an existing \( AUC \) predictive model which requires multiple sample points, we checked the predictive performance for our model. As the existing model for the \( AUC \), we used the method of Vathsala et al.,\(^{17}\) because they used the trough level of CsA for their \( AUC \) predictive model. Their model, which requires 3 sample points, is defined as follows:

\[
AUC_{0-12} = 4.44 \times C(0 \text{h}) + 2.42 \times C(2 \text{h}) + 5.91 \times C(6 \text{h}) + 83,
\]

where \( AUC_{0-12} \) (ng·h/ml) represents a product of the \( AUC \) integrated from time zero to 12 h after oral administration, and \( C(t) \) represents the CsA concentration in whole blood at trough (0 h) or 2 h and 6 h after the oral administration of CsA. As the \( AUC_{0-12} \) predictive model by Vathsala et al. permits the use of CsA measurements by the m-FPIA, our data from 48 time courses were applied to this equation, and the CSS was calculated by dividing the predicted \( AUC_{0-12} \) by 12 h. As described in the method of Shiner et al.,\(^{18}\) the bias, accuracy, and scattering in the CSS derived from ours and other predictive models were evaluated by calculating the mean prediction error (ME), mean absolute prediction error (MAE), and root mean squared prediction error (RMSE), respectively.

**Clinical Adaptation** To check the clinical validity of our \( AUC_{0-8} \) predictive model, we calculated the CSS in other patients with different clinical episodes. The trough levels after leaving the hospital were obtained when they attend as outpatients with intervals from 11.5 to 13.8 h after the last dose.

**RESULTS**

Figure 1 shows 48 clinical time-courses obtained from 13 renal transplant patients during CsA trough level monitoring from April 1990 to March 1996. These data involved all data obtained during or after episodes of hepatotoxicity, acute or chronic renal rejection, nephrotoxicity, acute tubular necrosis, gastroenteritis or anemia. There was great inter- and intra-individual variation in the shape of the time-courses. The \( AUC_{0-8} \) during nephrotoxicity and acute tubular necrosis, after acute renal rejection, or during gastrointestinal dysfunction were relatively reduced in comparison with that during no episodes.

The final results of multiple regression analysis for creating the \( AUC_{0-8} \) predictive model, and the predictive equation for the \( AUC_{0-8} \), are shown in Table 2. Assessed by the AIC, the logarithmic expression of the TL and OB are more closely correlated with the \( AUC_{0-8} \). There was no multicollinearity between each of the independent variables. The \( AUC_{0-8} \) was calculated by dividing the predicted \( AUC_{0-8} \) by 8 h.

**Table 2. Final Results in Creating an \( AUC_{0-8} \) Predictive Model Using 48 Clinical Time Courses in Renal Transplant Patients**

<table>
<thead>
<tr>
<th>Independent variables</th>
<th>Unit</th>
<th>Coefficient</th>
<th>Standardized coefficient</th>
<th>Partial correlation</th>
<th>VIF</th>
</tr>
</thead>
<tbody>
<tr>
<td>log(TL)</td>
<td>ng/ml</td>
<td>5673.1</td>
<td>0.689</td>
<td>0.823</td>
<td>1.23</td>
</tr>
<tr>
<td>log(OB)</td>
<td>%</td>
<td>9342.8</td>
<td>0.278</td>
<td>0.529</td>
<td>1.97</td>
</tr>
<tr>
<td>D&lt;sub&gt;pot&lt;/sub&gt;</td>
<td>mg/d</td>
<td>64.1</td>
<td>0.377</td>
<td>0.460</td>
<td>1.56</td>
</tr>
<tr>
<td>D&lt;sub&gt;TK&lt;/sub&gt;</td>
<td></td>
<td>869.4</td>
<td>0.224</td>
<td>0.317</td>
<td>2.43</td>
</tr>
<tr>
<td>HCT</td>
<td>%</td>
<td>-168.9</td>
<td>-0.387</td>
<td>-0.337</td>
<td>1.99</td>
</tr>
<tr>
<td>SG&lt;sub&gt;r&lt;/sub&gt;</td>
<td>mg/dl</td>
<td>-161.2</td>
<td>-0.283</td>
<td>-0.390</td>
<td>2.41</td>
</tr>
<tr>
<td>GPT</td>
<td>IU/l</td>
<td>-11.3</td>
<td>-0.183</td>
<td>-0.340</td>
<td>1.39</td>
</tr>
<tr>
<td>PL&lt;sub&gt;pot&lt;/sub&gt;</td>
<td>mg/dl</td>
<td>3.0</td>
<td>0.092</td>
<td>0.185</td>
<td>1.28</td>
</tr>
<tr>
<td>SEY&lt;sup&gt;+&lt;/sup&gt;</td>
<td></td>
<td>-588.6</td>
<td>-0.093</td>
<td>-0.165</td>
<td>1.66</td>
</tr>
<tr>
<td>Constant</td>
<td></td>
<td>-24794.5</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Not in the equation

\[
\begin{align*}
\text{DAT} & = d \\
\text{D<sub>pot</sub>} & = \text{mg/kg/d} \\
\text{AG} & = \text{years}
\end{align*}
\]

\( AUC_{0-8} = 5673.1 \times \log(\text{TL}) + 9342.8 \times \log(\text{OB}) + 64.1 \times D_{\text{pot}} + 869.4 \times D_{\text{TK}} - 168.9 \times HCT - 161.2 \times SG_{r} - 11.3 \times GPT + 3.0 \times PL_{\text{pot}} - 588.6 \times SEY - 24794.5,
\]

\( a) \) The values were given as binary data as follows: male or cadaveric donor, 1; female or living-related donor, 2. \( b) \) PL represents the sum of triglyceride and total cholesterol in plasma. The statistical significance for the multiple regression was \( p < 0.00001 (R^2 = 0.862, n = 48) \).
variables as assessed by the VIF because the values of VIF for each variable was less than 10. Assessed by the standardized coefficient, the contribution of variables made to the AUC_{0-8} was in the order log(TL) > HCT > D_{ord} > SCr > log(OB) > DTK > GPT > SEX > PL. The AUC_{0-8} shows positive correlation with log(TL), log(OB), D_{ord}, DTK, and PL, but negative correlation with HCT, SCr, GPT, and SEX. The statistical significance for the multiple regression of the AUC_{0-8} predictive model was $p<0.00001 (R^2=0.862, n=48)$ with the minimum AIC. However, the DAT and the AG did not correlate with the AUC_{0-8}. Furthermore, to our surprise, the D_{ord} also did not correlate with the AUC_{0-8}. If the DAT, AG and D_{ord} were entrapped into the regression as independent variables, the AIC for the regression was increased. Figure 2 shows the relationship between the actual AUC_{0-8} and the AUC_{0-8} predicted by our model. The regression equation between the actual AUC_{0-8} and the predicted AUC_{0-8} was $y = 0.897x + 298.31$ ($r^2 = 0.862$, $p<0.00001, n=48$). The statistical results by our predictive model suggest that about 86% of the variance in the AUC_{0-8} can be explained by the independent variables shown in Table 2.

Figure 3 shows scatter diagrams of actual C_{ss} and predicted C_{ss} by either ours or the existing AUC predictive model. Preliminarily, we found that the value of AUC_{0-8} was 1.1-fold larger than that of AUC_{0-12} using 12 cases of time course, from time zero to 12h after administration. Therefore, we adjusted the actual value for the model of Vathsala et al. by multiplying the actual C_{ss} of our data by a constant 0.9. The regression equation between the actual and estimated C_{ss} by the model of Vathsala et al. (see Materials and Methods) was $y = 0.839x + 40.454$ ($r^2 = 0.868$, $p<0.00001, n=48$), while that of our model was $y = 0.881x + 51.446$ ($r^2 = 0.857$, $p<0.00001, n=48$). The ME, AME and RMSE by our model were -11.89, 68.45, and 87.77, respectively, while those by another model were -35.29, 63.74, and 86.87, respectively. These results suggests that the predictive performance of our model is almost equal to that of Vathsala et al.

Table 3 shows the clinical adaptation of the AUC_{0-8} predictive model to other patients with different clinical episodes. For each patient, the C_{ss} was calculated based on the predicted AUC_{0-8} using the trough level of CsA, biochemical tests, a daily dose of PRD, and the patient’s basic information. The patients’ renal functions after transplantation were satisfactorily recovered, and they were discharged at the latest on day 60. Patient T.S. had no episodes during the indicated monitoring period (from day 64 to day 505 after renal transplantation). Patient Y.N. had acute gastroenteritis from day 159 after renal transplantation, followed by acute renal rejection (from day 163 to day 216). Patient K.M. had nephrotoxicity from day 133 to day 300 after renal transplantation, with a persistent increase in SCr (from 1.55 mg/dl to 1.97 mg/dl) without fever. Patient Y.O. essentially had no clinical episodes after renal transplantation; however, he had lower HCT values, from 25% to 30% lower, due to his complete gastrectomy in the past. The mean values of CsA-EP were increased in patient Y.N. and Y.O., each of whom had episodes of acute renal rejection or anemia due to a lower value of HCT. The trough levels of CsA for these patients were within 100 ng/ml to 200 ng/ml, that is to say, they were within a normal therapeutic range. However, the C_{ss} based on the AUC_{0-8} predictive model for patient K.M. showed a 2-fold increase in comparison with that of patient T.S.

![Predicted Performance of Our AUC_{0-8} Predictive Model in Comparison with An Existing AUC Predictive Model](image)

The predicted C_{ss} by our model was calculated by dividing a predicted AUC_{0-8} by 8h, while the predicted C_{ss} by an existing AUC predictive model of Vathsala et al. (see text) was calculated by dividing a predicted AUC_{0-12} by 12h. The observed C_{ss} for our model was calculated by dividing the observed AUC_{0-8} by 8h, and the observed C_{ss} for another model was then given by multiplying it by a constant 0.9. The regression equation between observed and predicted C_{ss} by our model was $y = 0.881x + 51.446$ (line a, $r^2 = 0.857$, $p<0.00001, n=48$), while that by Vathsala et al. (line b) was $y = 0.839x + 40.454$ (line b, $r^2 = 0.868$, $p<0.00001, n=48$).
DISCUSSION

Although routine monitoring of CsA trough levels is advocated following transplantation, it is often difficult to adjust the CsA dose to correspond to changes in particular disease states. In clinical practice, the concentration of CsA is measured only in a single blood concentration (tough level), however the tough level has shown to be a poor guide for an optimization of the CsA regimen in which the results of trough levels play a crucial role. Therefore, there is general acceptance that the AUC monitoring of CsA is superior to the trough level monitoring to adjust the CsA dosage and to obtain the target concentration. The use of the concept of AUC monitoring, however, results in painful transplants in clinical practice, because that technique requires multiple blood collection after oral administration.

To effectively adapt the results of CsA trough level monitoring to clinical practice, we examined the relationships between the distribution property of CsA in blood and disease states after renal transplantation, then derived a predictive model for the CsA-EP. The CsA-EP is given as a function of the results of CsA trough levels and biochemical tests which are measured at routine monitoring. This indicator reflects disease states after renal transplantation and CsA oral clearance, and it correlates with lymphocyte proliferation. As the present study is descended from our series of investigations, the AUC_{0-8} predictive model derived here also is given as a function of several routine monitoring results, namely, CsA trough levels, biochemical tests, a daily dose of PRD, and the patient’s basic information.

Several reports have demonstrated the clinical utility of AUC monitoring of CsA in renal transplant patients. Grel et al. concluded that the AUC but not the trough level was significantly correlated with the administered oral dose, expressed as the total mg (AUC: \( r = 0.381, p = 0.001 \); trough level: \( r = 0.151, p = 0.2 \)) or as milligrams per kilogram body weight (AUC: \( r = 0.538, p = 0.0001 \); trough level: \( r = 0.136, p = 0.26 \). On the other hand, Frey et al. reported that when the same dose of CsA was given to renal transplant patients, the tough levels correlated to the corresponding AUC. In our data, the trough levels of CsA did not correlate with daily oral dose expressed as milligrams per kilogram body weight (\( r = 0.019, n = 48, NS \)). There is some controversy in these observations; however, it is considered that the different sample matrices or assay methods may provide different results. The assay method and sample matrix used in the former report were serum and polyclonal radio immunoassay, and in the later report were whole blood and a specific technique such as monoclonal radio immunoassay or an HPLC. In this study, we measured the CsA concentration in whole blood by means of a specific method, said to be the m-FPIA; therefore, the trough level in the multiple regression was correlated with the AUC_{0-8}.

The data of time-courses used here were not intentionally collected throughout the monitoring period; therefore, the effects of intra-individual variability found in independent variables on the multiple regression were not strictly considered in creating the AUC_{0-8} predictive model. Although the effects of intra-individual variability may be in variance of regression coefficients or in the intercept, higher statistical significance for the regression suggests that the AUC_{0-8} predictive model created has universal validity, even though intra-individual variability exists. Creating the AUC_{0-8} predictive model, we selected obesity and a daily dose of PRD as predictors for the AUC_{0-8}. In our previous study, a higher SCR and elevated trough levels were found in psoriasis patients with higher obesity, even though they received almost the same CsA dose compared to psoriasis patients with lower obesity. The obesity, therefore, is a factor which influences CsA pharmacokinetics in renal transplant patients, and the AUC_{0-8} increases with a rise in the OB. However, it should be noted that in renal transplants an increase in body weight is not strictly distinguished between being due to an increase in body fat or due to systemic edema. In addition, a co-administrated drug, PRD, a known inducer of CYP 3A4 in liver microsomes, has been expected to have a drug interaction with CsA because CsA is metabolized by CYP3A4. Essentially, it is expected that the trough level of CsA will be reduced when CsA is co-administered with PRD. However, in clinical practice, several reports demonstrated that the trough level of CsA was actually increased by PRD. The detailed
mechanism of drug interaction between CsA and PRD was unknown; however, our $AUC_{0-8}$ predictive model clearly shows a drug interaction between the two because the $AUC_{0-8}$ has a positive correlation with the daily dose of PRD.

The rest, the fact that dose of CsA had no correlation with the $AUC_{0-8}$, supports that the pharmacokinetics of CsA follows a nonlinear kinetics.\(^3\)\(^,\)\(^2\(^5\)\) Moreover, a negative correlation between the $AUC_{0-8}$ and the HCT, the SCr, or the GPT means that the $AUC_{0-8}$ is increased during anemia, but decreased during renal dysfunction and hepatotoxicity. These observations agree with other clinical results of CsA pharmacokinetics which have been reported elsewhere.\(^3\)\(^,\)\(^2\(^6\)\) In addition, sex distinction or donor type of kidney are also influencing factors for CsA pharmacokinetics: a male or living-related donor increases the $AUC_{0-8}$, whereas a female or cadaveric donor decreases the $AUC_{0-8}$. The fact that the donor type of a transplanted kidney dominates the CsA pharmacokinetics is very interesting for us as a blind spot, because the therapeutic window for both types of donors is now treated as the same. Further investigation should be required regarding the effects of cadaverous or living-related donors on CsA pharmacokinetics.

There are several methods available to estimate the $AUC$ in addition to trapezoidal approximation. These methods also employ the multiple regression equation using several data points, including trough level after oral administration.\(^8\)\(^,\)\(^1\(^7\)\(^,\)\(^2\(^7\)\) However, in either case, multiple sample points are required when these methods are performed. In contrast, our $AUC_{0-8}$ predictive model uses routine trough levels and biochemical tests, a patient’s basic information, and a daily dose of PRD; therefore, this model reduces the pain for patients when the $AUC$ monitoring is performed. The predictive performance of our model was almost the same as that of the existing model using multiple data points after the oral administration of CsA, suggesting that our predictive model for the $AUC_{0-8}$ is convenient and useful in clinical practice.

In our institution, at the early phase of immunosuppressive therapy with CsA, within 30 d, we venture to maintain the CsA trough levels at a range of 150 ng/ml to 250 ng/ml; thereafter, during the maintenance phase, we venture to maintain a level ranging from 80 ng/ml to 150 ng/ml (therapeutic guide line). The trough levels for all patients enrolled in the clinical adaptation study of our $AUC$ predictive model during hospitalization were maintained within those therapeutic windows. Their renal functions were satisfactorily recovered during hospitalization, and they were discharged, at the latest, on day 60 after renal transplantation. Patients T.S. and Y.O. had no episodes throughout the monitoring period, but patient Y.O. had received a complete gastrectomy at the age of 24. Patient Y.N. had episodes of acute gastroenteritis on and after day 159, and thereafter, had an episode of acute renal rejection from day 163 to day 216. Patient K.M. had an episode of nephrotoxicity with a persistent increase in SCr on and after day 133. The mean trough levels of these patients listed in Table 3 were within the therapeutic range generally advocated. However, the estimates of $C_{\text{cr}}$ due to our $AUC_{0-8}$ predictive model, clearly distinguished the CsA concentration during disease states from that in patients with no episodes. In patient Y.N., before acute renal rejection, the estimated $C_{\text{cr}}$ was relatively lower than in patients with no episodes. In patient K.M., during nephrotoxicity, the estimated $C_{\text{cr}}$ was maintained at a relatively higher level. Grevel et al. reported that the target concentration of $C_{\text{cr}}$ with a specific monitoring technique was 370 ng/ml.\(^8\) In our patients, the range of $C_{\text{cr}}$ with no episodes was estimated to be between 200 ng/ml to 350 ng/ml. Taking differences in races into consideration, this range of $C_{\text{cr}}$ is the optimal target concentration of CsA at a steady-state in Japanese renal transplant patients.

The CsA-EP is an intensive parameter that depends on the trough level of CsA and on a patient’s disease state.\(^1\(^0\)\) An increase in the CsA-EP, therefore, means a lower bioavailability of CsA and being in the state of renal dysfunction, hepatotoxicity, or anemia.\(^9\)\(^-\)\(^1\(^2\)\) As found in patient K.M., who exhibited chronic nephrotoxicity without increases in the CsA-EP and trough level, cases with a relative 2-fold increase in the CsA despite the trough levels being within normal range, also existed. In this perspective, the CsA monitoring due to our $AUC_{0-8}$ predictive model is useful for determining subconscious nephrotoxicity.

In conclusion, our predictive model for the $AUC_{0-8}$ using routine trough levels of CsA, biochemical tests, a patient’s basic information, and a daily dose of PRD as predictors was satisfactory for the clinical use. Since our predictive model requires only trough level measurement of CsA, the use of this model for therapeutic drug monitoring of CsA will be connected with a cost benefit and also better patient comfort. However, it should be noted that the model is applicable to renal transplant patients who receive CsA with PRD, and that monitoring results at trough level in the whole blood by the m-FPIA must be used. An intentional use of our $AUC_{0-8}$ Predictive model as a protocol of CsA immunosuppressive therapy will provide an improvement in therapeutic control.

REFERENCES AND NOTES