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The relative importance of cyclosporine exposure in heart, kidney or liver transplant recipients on maintenance therapy

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Abstract We investigated the relationship between cyclosporine exposure and the presence of cyclosporine-related side effects and assessed the advantage of the cyclosporine concentration 2 h post-dose (C_2) over pre-dose concentration (C_0) monitoring. Cyclosporine area-under-the-concentration–time curves were measured during the absorption phase ($AUC_{0-4\,h}$) in 49 liver, 28 heart and 26 kidney transplant recipients (time since transplantation >6 years) with or without cyclosporine-related side effects on maintenance therapy. The cyclosporine C_0 correlated well with AUC_{0-4} ($r=0.77$), whereas C_2 levels correlated strongly with AUC_{0-4} ($r=0.92$). Although we observed a trend towards higher CsA concentrations in transplant recipients with side effects than in patients without CsA toxicity, the large majority of those differences were not statistically significant. Thus, as cyclosporine exposure was not clearly related to the presence of side effects, and C_0 correlated fairly with AUC_{0-4}, the advantage of monitoring cyclosporine treatment using C_2 rather than C_0, may be limited for patients on cyclosporine maintenance therapy.

Keywords Cyclosporine · Pharmacokinetics · Solid organ transplantation · Side effects · Toxicity · Therapeutic drug monitoring

Abbreviations *AUC*: Area under the concentration vs time curve · *C_0*: Pre-dose concentration (trough concentration) · *C_2*: Concentration 2 h post-dose · *C_{max}*: Peak concentration · *CN*: Calcineurin · *CsA*: Cyclosporine A · *TDM*: Therapeutic drug monitoring · *T_{max}*: Time-to-peak concentration · *TRL*: Tacrolimus

Introduction

The introduction of cyclosporine (CsA) in the early 1980s resulted in a significant improvement in the results of solid organ transplantation [1]. However, the clinical use of CsA is complex, due to its narrow therapeutic index, many drug interactions and highly variable pharmacokinetics [2]. Moreover, CsA has numerous side effects, such as nephrotoxicity, hypertension, hypercholesterolaemia and the induction of glucose intolerance [1]. Most transplantation centres have adopted the strategy of monitoring CsA using whole blood, pre-dose or trough

concentration (C_0) measurements and adjusting the CsA dose to reach a certain predefined C_0 target range, the limits of which may differ, depending on the organ transplanted and the time since transplantation [3].

The clinical utility of this approach suffers from the fact that, in de novo transplant recipients, the CsA C_0 does not predict total drug exposure over a 12 h or 24 h time period [as measured by the area-under the CsA concentration vs time curve (AUC)] at the individual level and does not correlate well with clinical outcome [4, 5, 6, 7, 8]. This is explained by the highly variable first-pass metabolism of CsA that occurs mostly during the first 4 h following oral administration of the drug [9]. Therefore, a potential risk of the monitoring of CsA using C_0 is that low drug exposure may not be detected, possibly resulting in under-immunosuppression and the risk of acute rejection.

Likewise, high CsA exposure may go unnoticed, resulting in (long-term) toxicity. Some have, therefore, advocated the use of an abbreviated AUC instead of the C_0 to monitor CsA therapy. The CsA AUC in the first 4 h after oral administration (AUC_{0-4}) has been shown to correlate well with the AUC_{0-12} and to predict clinical outcome after kidney transplantation [5, 6, 10]. Because the determination of an AUC_{0-4} is time consuming, expensive and not practicable for use in an outpatient clinic, there has been continuing interest in simpler parameters for therapeutic drug monitoring (TDM) of CsA. The whole-blood CsA concentration 2 h after administration of the drug (C_2) was shown to be the single time point with the best correlation with total drug exposure [6, 9, 11].

Subsequently, C_2 monitoring has been used for TDM in several clinical trials and has, generally, resulted in a low or decreased incidence of acute rejection and excellent (renal) tolerability when compared with C_0 monitoring [12, 13, 14, 15, 16]. Following the outcomes of these trials, target values for C_2 have been identified. Currently, the recommended C_2 values for liver and kidney transplant recipients more than 6 months after transplantation are 600 ng/ml ± 20% and 800 ng/ml ± 20%, respectively [9, 11, 16]. C_2 target levels have not yet been established for heart transplant recipients. The measuring of C_2 concentrations, however, does require a considerable effort to reliably draw blood at exactly the correct time point. Because of the practical limitations of this approach, many transplantation centres have not changed their policy of performing TDM on the basis of CsA trough levels.

Although the correlation between acute rejection and nephrotoxicity and CsA exposure as measured by an AUC_{0-4} or C_2 has been established in de novo transplant recipients, the relation between drug exposure and other CsA-related side effects is less clearly defined, especially in patients on long-term CsA treatment. We feel that this is very important because these other CsA-related side effects negatively influence patient survival, quality of life and the long-term outcome after transplantation. Moreover, the occurrence of CsA-related side effects may lead to patient non-compliance, with the risk of acute rejection. In our centre we routinely measure CsA C_0 after kidney, liver and heart transplantation. We do acknowledge that individual patients may suffer from side effects that seem to be CsA related, although predose concentrations are within, or even at the lower end of, the defined target range. Possibly, the use of another method for TDM, i.e. an AUC_{0-4} or C_2, would recognize the increased exposure to CsA in these patients.

The aim of this study was twofold. First we investigated whether solid organ allograft recipients with CsA-related side effects on maintenance therapy with CsA and with CsA C_0 within the therapeutic range, had a higher exposure to CsA than did a control group of transplant recipients, at similar CsA C_0 but without CsA-related side effects. We therefore measured the CsA AUC_{0-4} in 103 liver, kidney and heart allograft recipients more than 6 months after transplantation. Second we determined how many of those patients had C_2 levels above the currently recommended target ranges.

Materials and methods

Patients

During routine outpatient clinical visits, all patients who had received a heart, kidney or liver transplant at the Erasmus Medical Center in The Netherlands were asked to participate in the study. Patients had to have been on CsA treatment, for at least 3 months without changes in CsA dosage, during the 3 months before entry into the study. All patients used the CsA micro-emulsion formulation (Neoral, Novartis) twice daily in two equally divided doses. Patients taking medication known to interact with CsA, such as the calcium-channel blockers diltiazem, nicardipine or verapamil, anti-epileptics (phenytoin and carbamazepine), anti-mycotics (fluconazole and ketoconazole) and macrolide antibiotics (erythromycin and clarithromycin), were not included in the study.

On the day of the pharmacokinetic study, patients were (physically) examined for the presence of renal insufficiency (serum creatinine ≥125 μmol/l in liver and heart transplant recipients; not determined in kidney transplant recipients), hypertension (blood pressure ≥150/100 mmHg or the use of antihypertensive medication), hypercholesterolaemia (total serum cholesterol > 7.5 mmol/l or the need for lipid-lowering drugs that was not present prior to transplantation), gum hyperplasia, hirsutism and hypertrichosis, polyneuropathy or tremor of the hands (not caused by diabetes mellitus or otherwise explained by co-medication such as theoph-

ylline or sympathicomimetics), diabetes mellitus (defined by the need for glucose-lowering drugs that was not present before transplantation) and (post-transplantation) gout. If any of these symptoms was present, on the day of the study as well as during the 3-month period before entry into the study (determined by history taking and patient chart review), patients were classified as having CsA-related side effects. As a control group we selected solid organ allograft recipients who exhibited none of the above-mentioned CsA-related side effects. The study was carried out in accordance with the declaration of Helsinki and was approved by the ethics committee of the Erasmus Medical Center. All patients gave written informed consent.

Cyclosporine AUC_{0-4} measurement

On the day of the AUC_{0-4} measurement an intravenous cannula was inserted and maintained with 0.9% NaCl solution. After the C_0 whole-blood sample had been drawn, patients were asked to take their CsA. Following CsA administration, blood was drawn at 1, 2, 3 and 4 h. All patients had been instructed to take their regular CsA dose 12 h before, on the previous day. The blood samples were frozen and stored at −30°C until CsA concentration was determined. CsA concentrations were determined by Emit 2000 assay (Syva, Dade Behring, Cupertino, Calif., USA) on a Cobas Mira Plus analyser (Roche). We used the trapezoidal rule to calculate the AUC_{0-4}. The peak CsA concentration (C_{max}) and the time to peak CsA concentration (T_{max}) were obtained directly from the data.

Statistical analysis

We used Student's unpaired *t*-test, Fisher's exact test with Yates' continuity correction or one-way ANOVA followed by Tukey's post-hoc test, as appropriate, to compare pharmacokinetic parameters. For correlation analysis, we calculated Pearson's correlation coefficient, followed by linear regression and used Fisher's Z-transformation to compare correlation coefficients. Unless stated otherwise, data are presented as means ± SD. A *P* value of less than 0.05 was considered statistically significant.

Results

General data

A total number of 103 patients was included, of whom 49 had received a liver transplant, 28 a heart transplant and 26 patients a kidney transplant. The mean age at the time of transplantation was 45.7 ± 11.4 years for liver transplant recipients, 45.7 ± 14.3 years for heart transplant recipients and 46.2 ± 14.4 years for kidney transplant recipients and was not different between the three groups ($P = 0.99$, one-way ANOVA). Time after transplantation was comparable between the three groups, with a mean follow-up time of 6.3 ± 3.1 years for liver transplant recipients, 6.7 ± 3.7 years for heart transplant recipients and 7.2 ± 5.9 years for kidney transplant recipients ($P = 0.63$). The other patient characteristics are summarized in Table 1.

CsA pharmacokinetics

Daily CsA dose was comparable between all three groups: 216 ± 80 vs 229 ± 76 vs 238 ± 61 mg/day for liver, heart and kidney allograft recipients, respectively ($P = 0.44$; Table 2). CsA dose, calculated on a milligramme per kilogramme bodyweight basis, was equal in all three groups as well: 2.9 ± 1.2 vs 2.9 ± 1.0 vs 3.1 ± 0.9 mg/kg/day for liver, heart and kidney allograft recipients, respectively ($P = 0.79$; Table 2). However, liver transplant recipients were maintained at significantly lower CsA pre-dose concentrations than were kidney transplant recipients: 115 ± 46 vs 144 ± 50 ng/ml ($P < 0.05$). The mean CsA C_0 of heart transplant recipients was not significantly different from those observed in either liver or kidney allograft recipients. In addition, CsA C_1, C_2, C_3, AUC_{0-4} and C_{max} were all significantly lower in liver transplant recipients than in kidney allograft recipients, but not heart transplant recipients (Table 2). As illustrated in Fig. 1, C_0 correlated fairly with AUC_{0-4} and numerically less well with C_2: Pearson's r (r^2) 0.77 and 0.72 (0.59 and 0.51), respectively ($P < 0.0005$). However, this difference was not statistically significant ($P = 0.42$, comparison of Pearson's correlation coefficients using Fisher's Z-transformation). The correlation between C_2 and AUC_{0-4} was strong, with an r (r^2) of 0.92 (0.85; $P < 0.0005$). The difference in correlation of C_0 and C_2 with AUC_{0-4} was statistically significant ($P < 0.0005$).

CsA pharmacokinetics and CsA-related side effects

Of the 49 liver transplant recipients included in the study, 30 (61.2%) had CsA-related side effects, whereas 19 (38.8%) had none. Hypertension was present in 25 of the 30 patients with side effects (51.0%), renal insufficiency in 22 patients (44.9%) and hypertrichosis/hirsutism in 21 patients (42.9%). Gingival hyperplasia was found in 11 patients (22.4%), and tremor of the hands and hypercholesterolaemia were each found in seven

Table 1 Characteristics of 103 solid organ transplant recipients. All values are expressed as means ± SD

Characteristic	Liver	Heart	Kidney
Number of patients	49	28	26
Male/female (*n*)	20/29	22/6	17/9
Age at time of transplantation (years)	45.7 ± 11.4	45.7 ± 14.3	46.2 ± 14.4
Range (years)	19–64	14–66	13–71
Time since transplantation (years)	6.3 ± 3.1	6.7 ± 3.7	7.2 ± 5.9
Range (years)	1.4–12.6	0.9–15.3	1.6–26
Underlying disease			
Primary sclerosing cholangitis	8		
Primary biliary cirrhosis	10		
Hepatitis B	8		
Hepatitis C	2		
Alcohol-induced liver cirrhosis	5		
Acute or toxic liver failure	7		
Unknown cause	5		
Other	4		
Ischaemic heart disease		13	
Cardiomyopathy		14	
Congenital heart disease		1	
Hypertensive nephropathy			6
Autosomal dominant polycystic kidney disease			4
Diabetic nephropathy			1
Glomerulonephritis			2
Unknown cause			8
Other			5

Table 2 CsA dosage and pharmacokinetics in three groups of solid organ transplant recipients. All values are expressed as means ± SD. *NS* not significant

Transplantation type	Liver	Heart	Kidney	P^a
Number of patients	49	28	26	
CsA dose (mg/day)	216 ± 80	229 ± 76	238 ± 61	NS
CsA dose (mg/kg per day)	2.9 ± 1.2	2.9 ± 1.0	3.1 ± 0.9	NS
CsA C_0(ng/ml)	115 ± 46	122 ± 50	144 ± 50	< 0.05
CsA C_1(ng/ml)	754 ± 350	926 ± 352	1014 ± 452	< 0.05
CsA C_2(ng/ml)	641 ± 248	729 ± 290	884 ± 191	< 0.001
CsA C_3(ng/ml)	434 ± 202	449 ± 194	556 ± 160	< 0.05
CsA C_4(ng/ml)	305 ± 143	312 ± 140	382 ± 112	NS
CsA AUC_{0-4}(ng/ml per h)	2,039 ± 727	2321 ± 827	2,718 ± 671	< 0.01
C_{max}(ng/ml)	833 ± 319	975 ± 303	1,116 ± 365	< 0.01
T_{max}(h)	1.4 ± 0.6	1.3 ± 0.4	1.4 ± 0.5	NS

[a] *P* values indicate differences in pharmacokinetic parameters between liver transplant recipients and kidney transplant recipients (one-way ANOVA with Tukey's post-hoc test)

patients (14.3%). No liver transplant recipients with gout were identified. Although there was an overall trend towards lower CsA concentrations in patients without side effects than in the group of patients with CsA toxicity, none of these differences reached statistical significance (Table 3).

Next, we compared CsA exposure in patients suffering from an individual side effect with that in patients who did not have that particular side effect (Table 4). Patients with gingival hyperplasia used significantly more CsA than did patients who did not have gingival hyperplasia: 3.6 ± 1.5 vs 2.7 ± 1.0 mg/kg per day, respectively ($P = 0.025$). As a result, CsA exposure (C_2, C_{max} and AUC_{0-4}) was also significantly higher in patients with gingival hyperplasia (Table 4). Likewise, patients with hypertrichosis or hirsutism used significantly more CsA than those patients with no excessive hair growth: 3.3 ± 1.5 vs 2.6 ± 0.8 mg/kg per day, respectively ($P = 0.045$). This difference was reflected by a higher CsA C_{max} in the former patient group: 936 ± 325 vs 755 ± 298 ng/ml ($P = 0.049$). For all other side effects studied (including nephrotoxicity and hypertension, data not shown) CsA exposure in liver transplant recipients with a specific side effect was not statistically, significantly different from patients who did not have that side effect (Table 4).

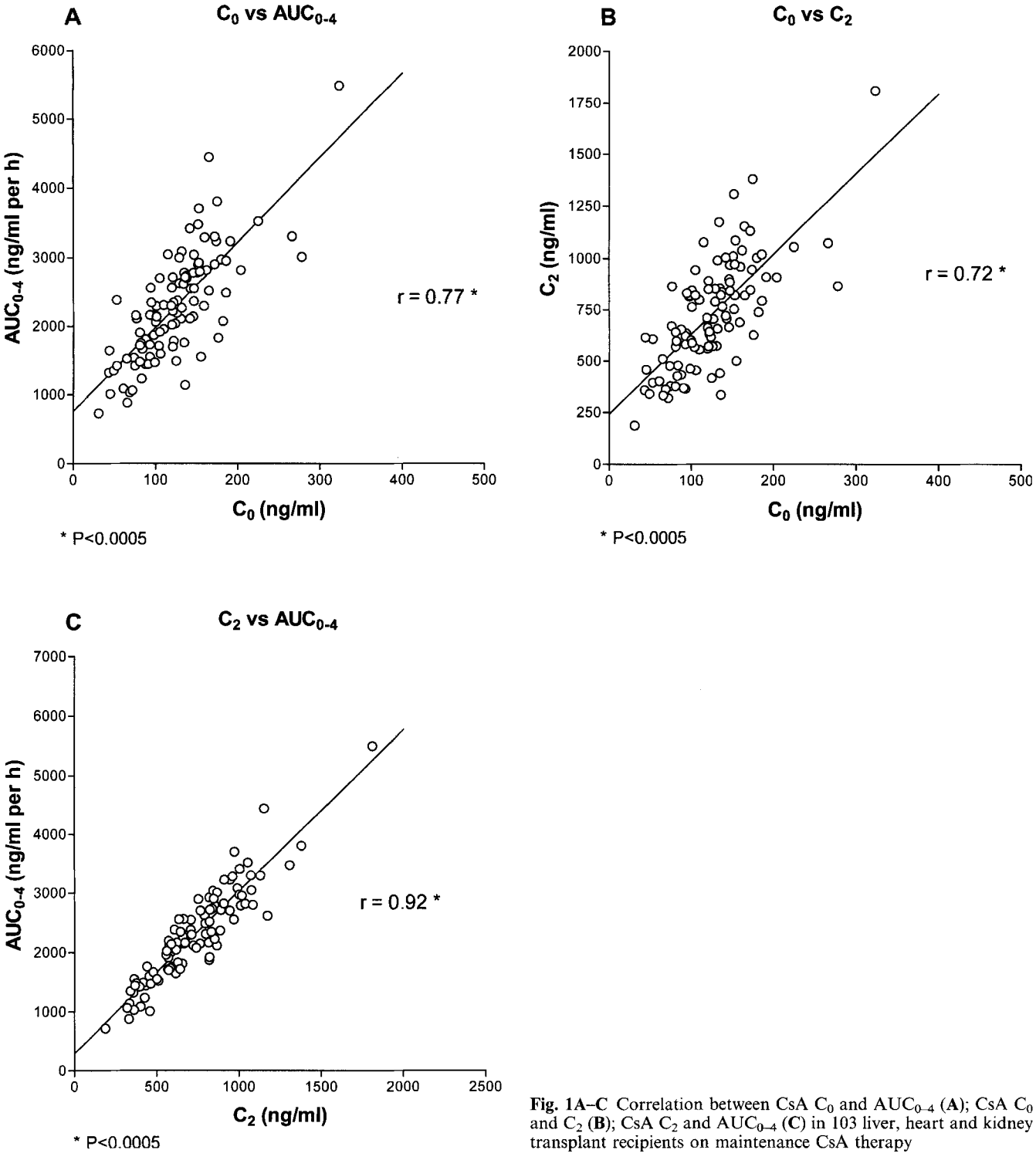

Fig. 1A–C Correlation between CsA C_0 and AUC_{0-4} (**A**); CsA C_0 and C_2 (**B**); CsA C_2 and AUC_{0-4} (**C**) in 103 liver, heart and kidney transplant recipients on maintenance CsA therapy

Of the 28 heart transplant recipients included in the study, 20 were identified as having side effects. Hypertension and hypertrichosis/hirsutism were the most common, each present in 18 (64.3%) patients, followed by hypercholesterolaemia (12 patients; 42.9%), renal insufficiency (11 patients; 39.3%), tremor/polyneuropathy (ten patients; 35.7%) and gingival hyperplasia (nine patients; 32.1%). When CsA pharmacokinetics were compared between heart transplant recipients with, and those without, any side effects, no significant differ-

Table 3 CsA-related side effects and CsA pharmacokinetics. No statistically significant differences in any of the CsA pharmacokinetic parameters were observed between patients with and patients without side effects in the three groups. All values are expressed as means ± SD

Parameter	Liver			Heart			Kidney		
	Side effects ($n=30$)	No side effects ($n=19$)	P	Side effects ($n=20$)	No side effects ($n=8$)	P	Side effects ($n=14$)	No side effects ($n=12$)	P
CsA dose (mg/day)	225 ± 92	201 ± 56	0.27	230 ± 83	225 ± 60	0.88	236 ± 60	242 ± 63	0.81
CsA dose (mg/kg per day)	3.0 ± 1.4	2.7 ± 0.8	0.32	2.9 ± 1.1	3.0 ± 1.0	0.87	3.0 ± 1.0	3.1 ± 0.7	0.85
CsA C_0(ng/ml)	121 ± 48	107 ± 43	0.33	122 ± 57	124 ± 25	0.90	147 ± 50	141 ± 51	0.74
CsA C_1(ng/ml)	814 ± 338	659 ± 357	0.13	943 ± 384	883 ± 273	0.69	1,072 ± 488	947 ± 416	0.49
CsA C_2 (ng/ml)	678 ± 246	582 ± 244	0.19	722 ± 318	747 ± 225	0.85	899 ± 208	868 ± 178	0.69
CsA C_3(ng/ml)	454 ± 215	403 ± 182	0.40	448 ± 217	451 ± 129	0.98	554 ± 170	559 ± 155	0.94
CsA C_4(ng/ml)	322 ± 160	277 ± 108	0.29	311 ± 159	316 ± 83	0.94	376 ± 106	388 ± 122	0.79
CsA AUC_{0-4}(ng/ml per h)	2,167 ± 710	1,837 ± 725	0.12	2,329 ± 943	2,300 ± 471	0.94	2,786 ± 751	2,638 ± 587	0.59
C_{max}(ng/ml)	895 ± 303	735 ± 328	0.09	977 ± 331	969 ± 236	0.95	1,147 ± 428	1,080 ± 288	0.65
T_{max}(h)	1.4 ± 0.6	1.5 ± 0.6	0.46	1.3 ± 0.4	1.3 ± 0.5	1.00	1.4 ± 0.5	1.4 ± 0.5	0.81

Table 4 CsA-related side effects and CsA pharmacokinetics in liver, heart and kidney transplant recipients. All values are expressed as means ± SD

Transplantation type	PK parameter	Gingival hyperplasia		Hypertrichosis/hirsutism		Tremor/polyneuropathy		Hypercholesterolaemia	
		Yes	No	Yes	No	Yes	No	Yes	No
Liver	n	11	38	21	22	7	22	7	42
	CsA dose (mg/kg per day)	3.6 ± 1.5	2.7 ± 1.0[a]	3.3 ± 1.5	2.6 ± 0.8[a]	3.4 ± 1.6	2.8 ± 1.1	3.2 ± 1.8	2.8 ± 1.1
	CsA C_0(ng/ml)	128 ± 46	112 ± 46	124 ± 50	109 ± 42	125 ± 49	114 ± 46	122 ± 57	114 ± 45
	CsA C_2(ng/ml)	775 ± 259	602 ± 233[a]	705 ± 252	592 ± 237	746 ± 190	623 ± 253	691 ± 229	632 ± 252
	CsA AUC_{0-4} (ng/ml per h)	2,514 ± 727	1901 ± 676[a]	2,251 ± 734	1,879 ± 692	2,443 ± 614	1,971 ± 729	2,299 ± 753	1,996 ± 723
	C_{max}(ng/ml)	1,054 ± 340	769 ± 287[b]	936 ± 325	755 ± 298[a]	1042 ± 315	798 ± 310	1,018 ± 336	802 ± 310
Heart	n	9	19	18	10	10	18	12	16
	CsA dose (mg/kg per day)	3.3 ± 1.2	2.8 ± 0.9	3.0 ± 1.0	2.8 ± 1.0	3.3 ± 1.2	2.7 ± 0.9	3.0 ± 1.1	2.9 ± 1.0
	CsA C_0(ng/ml)	145 ± 75	112 ± 29	126 ± 59	117 ± 29	128 ± 71	119 ± 34	133 ± 69	115 ± 28
	CsA C_2(ng/ml)	858 ± 407	668 ± 201	749 ± 324	694 ± 228	771 ± 394	706 ± 224	798 ± 387	678 ± 188
	CsA AUC_{0-4} (ng/ml per h)	2,744 ± 1178	2121 ± 526	2,415 ± 958	2,152 ± 520	2,478 ± 1143	2,234 ± 610	2,600 ± 1095	2,112 ± 494
	C_{max}(ng/ml)	1,098 ± 374	916 ± 253	1014 ± 328	904 ± 251	1,014 ± 357	953 ± 277	1,082 ± 345	894 ± 248
Kidney	n	11	15	11	15	7	19	4	22
	CsA dose (mg/kg per day)	3.1 ± 1.1	3.1 ± 0.7	3.0 ± 0.8	3.1 ± 1.0	3.6 ± 1.0	2.8 ± 0.7[a]	3.1 ± 0.8	3.1 ± 0.9
	CsA C_0(ng/ml)	142 ± 52	146 ± 49	152 ± 55	139 ± 47	157 ± 61	140 ± 46	155 ± 24	142 ± 53
	CsA C_2(ng/ml)	884 ± 148	885 ± 223	907 ± 204	868 ± 187	1,017 ± 201	835 ± 167[a]	1,086 ± 233	848 ± 163[a]
	CsA AUC_{0-4} (ng/ml per h)	2,789 ± 715	2,665 ± 657	2,816 ± 764	2,646 ± 612	3061 ± 810	2,591 ± 587	3,440 ± 938	2,587 ± 542[a]
	C_{max}(ng/ml)	1,187 ± 452	1,064 ± 290	1,163 ± 450	1,082 ± 300	1,233 ± 495	1,073 ± 309	1,500 ± 582	1,046 ± 277[a]

[a] $P < 0.05$
[b] $P < 0.01$

ences were observed in any pharmacokinetic parameter (Table 3). The pharmacokinetics of the patients suffering from individual side effects (including nephrotoxicity, hypertension and gout) were not different from those of patients without those individual side effects (Table 4).

Finally, CsA-related side effects were identified in 14 of the 26 renal transplant recipients included in the study. Hypertrichosis/hirsutism and gingival hyperplasia were the most frequently observed side effects and were each present in 11 patients (42.3%). Tremor or polyneuropathy was identified in seven patients (26.9%) and hypertension and hypercholesterolaemia were present in four patients (15.4%); one patient (3.8%) suffered from gout. As renal insufficiency in kidney transplant recipients is often determined by many fac-

tors and is difficult to distinguish from CsA nephrotoxicity (especially in the absence of a kidney biopsy), this side effect was not studied in this patient group. However, all patients that were classified as having no side effects did not show any clinical evidence for the presence of CsA nephrotoxicity. As in liver and heart transplant recipients, no significant differences in any of the CsA pharmacokinetic parameters were observed between the groups of patients with and without CsA side effects (Table 3). When individual side effects were analysed, we observed a higher CsA dose and CsA C_{max} in patients with neurotoxicity than in patients who did not have tremor or polyneuropathy: 3.6 ± 1.0 vs 2.8 ± 0.7 mg/kg per day ($P = 0.036$) and 1017 ± 201 vs 835 ± 167 ng/ml ($P = 0.028$), respectively. In addition, the four patients with hypercholesterolaemia had a higher CsA exposure than those patients with normal serum cholesterol levels (Table 4). CsA exposure in patients with gingival hyperplasia, hypertrichosis/hirsutism, hypertension or gout was comparable to that in patients without those specific side effects (Table 4).

CsA-related side effects and C_2

Of the 49 liver transplant recipients, 19 (38.8%), had a C_2 value above the recommended target range of 600 ng/ml ± 20%. C_2 was below this target range in 16 (32.7%) liver transplant recipients (Fig. 2A). The percentage of patients with C_2 levels above target was not different between the group of liver transplant recipients with or without cyclosporine-related side effects ($P = 0.55$, Fisher's exact test with Yates' continuity correction). Of the 26 kidney transplant recipients, eight (30.8%) had a C_2 value above the recommended target value of 800 ng/ml ± 20%. The C_2 value was below this target range in two (7.7%) kidney transplant recipients (Fig. 2B). Again, the number of patients with C_2 levels above target was not different between the group of kidney transplant recipients with or without cyclosporine-related side effects ($P = 1.00$). Of the 28 heart transplant recipients, ten (35.7%) had a C_2 value above 600 ng/ml ± 20%. In four of those patients (14.3%) the C_2 value exceeded 800 ng/ml ± 20%.

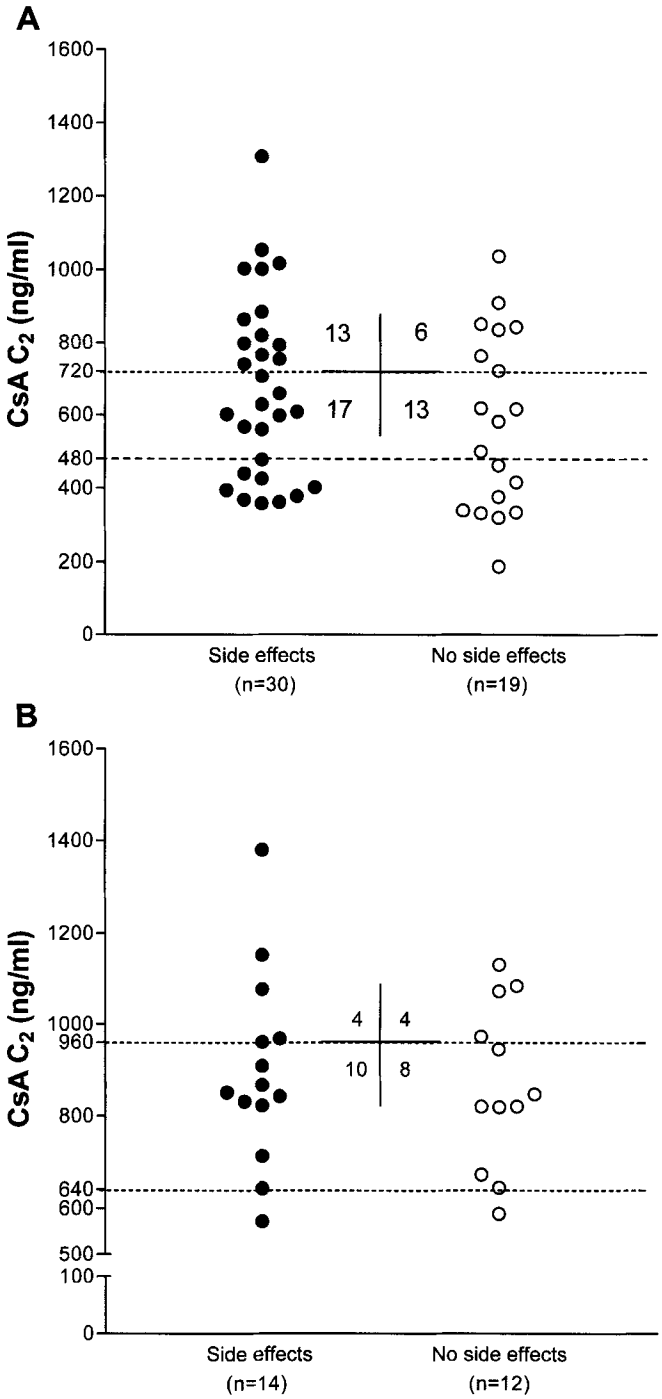

Fig. 2 CsA C_2 levels and CsA-related side effects in 49 liver (**A**) and 26 kidney (**B**) transplant recipients. The target level ± 20% range is indicated by *dotted lines*

Discussion

The introduction of micro-emulsified CsA and the publication of several clinical trials that compared the effectiveness of CsA to tacrolimus (TRL), have led to a renewed interest in TDM and the pharmacokinetics of CsA [17, 18]. In recent years, both C_2 and AUC_{0-4} have been demonstrated to be useful CsA-monitoring tools that correlate well with the incidence of acute rejection (AR) and nephrotoxicity [5, 6, 10, 12, 13, 14, 15]. However, most of those studies were performed in de novo transplant recipients and did not relate CsA pharmacokinetics to CsA-related side effects other than renal insufficiency or hypertension.

The kidney transplantation programme of the Erasmus Medical Center started in 1971 and was followed by the heart transplant programme in 1984 and the liver transplant programme in 1986. Since then, more than 1,500 kidney, 400 heart and 350 liver transplantations have been carried out. For many years CsA was the calcineurin inhibitor of choice, but, in recent years, we have switched to TRL-based immunosuppressive regimens for our kidney and liver transplant recipients. However, many of our patients still use CsA and often suffer from CsA-related side effects. In the present study we therefore investigated the relationship between CsA pharmacokinetics and CsA-related side effects.

In our cohort of patients on CsA maintenance therapy, CsA pharmacokinetics did not correlate well with the presence of CsA-related side effects. Although we did find an overall trend towards higher CsA exposure in patients with (a specific) side effect(s), the majority of those differences did not reach statistical significance. In addition, the same cyclosporine exposure that was associated with the presence of a particular side effect in one type of transplant recipient was not related to the occurrence of that same side effect in patients who had had a different organ transplanted. In our opinion, this argues against a clear relationship between CsA whole-blood concentrations and the presence of CsA toxicity.

David-Neto et al. studied a paediatric kidney transplant cohort and found statistically significant correlations between CsA pharmacokinetics and the occurrence of side effects [19]. An AUC greater than or equal to 4,158 ng/ml predicted the presence of hypertrichosis, whereas a C_{max} greater than or equal to 878 ng/ml was the best predictor for the appearance of tremors. Gingival hyperplasia was not associated with any of the pharmacokinetic parameters studied [19]. Those results are not necessarily contradictory to our findings. The mean CsA dose and AUC_{0-4} of our adult patients were much lower than those reported in the Brazilian paediatric study cohort. This could be explained by the fact that most of our patients were on long-term CsA therapy and many of them had already undergone several CsA dose reductions before the start of the study.

In many cases, the presence of side-effects had been an important reason for CsA dose reduction. Those previous dose reductions could have reduced a difference in CsA exposure that might have existed between patients with or without side effects. As this was a cross-sectional study we do not have data on CsA exposure at the time of emergence of CsA-related side effects. Furthermore, a positive selection of the investigated patients might have occurred, as patients with severe side effects might have been switched to CsA-free immunosuppressive regimens prior to the start of our study.

Our observations raise the question as to whether a further CsA dose reduction in our population will result in a decrease in the incidence and severity of side effects. CsA exerts its immunosuppressive effect through inhibition of calcineurin (CN), an enzyme that is important for the activation of T cells. Several studies investigating the relationship between CsA pharmacokinetics and CN inhibition demonstrated that, in vivo, CN is only partially inhibited. At C_0 (ranging between 148 and 180 ng/ml), CN was inhibited by 50%, while CsA peak concentrations (approximately 400 to 1800 ng/ml) resulted in around 70%–80% CN inhibition [20, 21, 22]. Moreover, CN inhibition rarely reached 100% and was greater in some tissues due to drug accumulation [22].

From these data it can be concluded that, at commonly used CsA target levels, the maximum pharmacodynamic effect of the drug is obtained and that further increasing drug blood levels will probably result only in a high incidence of side effects and considerable drug toxicity. Therefore, many transplant patients probably receive too much CsA and can undergo dose reduction, while the desired immunosuppressive effect of the drug is still maintained. More than one-third of our patients had C_2 levels above currently recommended target ranges, and adaptation of C_2 monitoring could result in (early) identification of CsA "overexposure" and, subsequently, in (further) dose reductions. Levy et al. recently reported the results of conversion of liver transplant patients in the maintenance phase from C_0 to C_2 monitoring [23]. Of the 351 patients that were converted, 36% had C_2 levels above the recommended target range. In those patients, a mean CsA dose reduction of 16% was required to achieve target range, resulting in a significant improvement of renal function, blood pressure and serum cholesterol [23].

Similar results have recently been reported for renal transplant recipients [24]. To study whether this approach will also lead to fewer (or less severe) side effects in our patient cohort, we are currently converting all liver transplant patients reported here to C_2 level monitoring followed by dose reduction, if indicated. However, C_0 correlated much better with AUC_{0-4} than has been reported previously [5, 24]. Possibly, the difference in time after transplantation explains the difference between the results of Mahalati et al. and our own. Nonetheless, our results may indicate that the reported beneficial effects of C_2 level monitoring might be limited for patients on CsA maintenance therapy. Lowering currently used C_0 target levels could result in a substantial CsA dose reduction as well, without the logistic problems associated with (the implementation of) C_2 level monitoring.

Alternatively, conversion of patients to TRL is another possibility to decrease the incidence and severity of CsA-related side effects. In 55 heart transplant recipients that were converted from CsA to TRL-based immunosuppressive therapy at our centre, a significant im-

provement in blood pressure, serum cholesterol and gum hyperplasia, without signs of acute rejection, was observed, even in patients who were as long as 14 years post-transplantation [25].

In kidney transplant recipients, conversion to TRL has been shown to be safe and to have resulted in lower serum cholesterol levels with an improvement in gingival hyperplasia and hypertrichosis [26, 27] with an improved creatinine clearance in one study [27]. In a retrospective analysis of 94 liver transplant recipients, converted to TRL for a variety of reasons, conversion resulted in a reduction of serum creatinine from 167 ± 36 to 119 ± 28 mmol/l (1 year after conversion) [28]. Besides conversion to TRL, complete cessation of CsA is another possibility that has been studied. The results of the meta-analysis by Kasiske et al. demonstrate that discontinuation of CsA results in an 11% higher risk for the development of acute rejection than in controls, in kidney transplantation [29]. However, the relative risk of graft failure was not significantly different from that of the control group.

In conclusion, we demonstrate no clear differences in CsA exposure in solid organ transplant recipients with or without CsA-related side effects. CsA C_2 levels were above currently recommended target ranges in 38.8% of liver and 30.8% of kidney transplant recipients, but C_2 levels above target were not more frequent in patients with side effects than in those with none. CsA dose reduction could be effective and safe in those patients. However, as the correlation between C_0 and AUC_{0-4} was better than that previously reported, the advantage of C_2 over conventional C_0 level monitoring might be limited in patients on low or moderate dose CsA maintenance therapy.

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