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ORIGINAL ARTICLE

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A pharmacokinetic comparison of the corn oil versus microemulsion gelcap formulation of cyclosporin used de novo after renal transplantation

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Abstract The initial poor absorption of the corn oil-based, gel capsule oral formulation of cyclosporin (CyA) greatly limits its use for inception of immunosuppressive therapy. Insufficient drug concentrations during the early post-transplant period predispose to renal allograft rejection. The present study served to compare the time required to achieve therapeutic CyA concentrations after de novo administration of the corn oil-based gel capsule (CyA-GC; $n = 11$) versus the microemulsion (CyA-ME; $n = 11$) formulation of CyA. During the 1st month after renal transplantation, patients underwent serial pharmacokinetic profiling from which we obtained observed and dose-corrected values of several parameters. Although patients in neither the CyA-GC nor the CyA-ME group adequately absorbed the drug during days 0–2, from day 3 to 4 patients in the CyA-ME group showed significantly greater absorption than those in the CyA-GC group ($P = 0.041$). Patients in the CyA-ME group reached the 1st month target average concentration (C_{av}) values (≥ 550 ng/ml) earlier than those in the CyA-GC group and required significantly lower daily CyA doses ($P = 0.018$). We conclude that therapeutic CyA levels can be achieved more rapidly and with lower doses of the drug after de novo administration of CyA-ME than with CyA-GC.

Key words Cyclosporin, microemulsion formulation · Microemulsion formulation, cyclosporin · Corn oil, cyclosporin

Introduction

The clinical application of cyclosporin (CyA) is complicated by tremendous intra- and inter-individual variations in drug absorption, distribution, metabolism, and elimination [1, 9, 12]. One major obstacle to achieving therapeutic drug concentrations is the low and variable oral bioavailability of the corn oil-based gel capsule (CyA-GC) or olive oil-based liquid Sandimmune formulations (Sandoz, Basel, Switzerland) of CyA. Although the bioavailability of CyA increases during the first 3 months after transplantation, low initial drug exposure presents a significant risk for renal allograft rejection [6]. The new microemulsion formulation of CyA (CyA-ME; Neoral, Sandoz), which is composed of a micellar system [5] containing a surfactant, lipophilic solvent, and hydrophilic solvent and co-solvent, enables CyA to be more rapidly dispersed in the intestine with less dependence upon the actions of bile and succus entericus [11]. Therefore, it is believed that therapeutic drug concentrations are achieved more quickly and consistently after administration of CyA-ME than after the oil-based formulations. In the present study, the time and the dose required to achieve therapeutic drug concentrations in the immediate post-transplant period (de novo) after CyA-GC versus CyA-ME administration were compared using serial pharmacokinetic profiles of renal transplant recipients.

Materials and methods

Immunosuppression

For all 22 patients, the initial CyA dose was selected on the basis of pretransplant test dose pharmacokinetic studies that were performed 2 weeks to 6 months before the transplant procedure after the sequential administration of intravenous, followed by oral, CyA [3]. Gel capsules were routinely used for pretransplant studies because CyA-ME was not available at the time of these studies. In the immediate postoperative period, no CyA was administered for 8–12 h. Thereafter, based on an open label randomization design, patients were assigned to receive either the CyA-GC or CyA-ME formulation at the selected initial CyA dose twice a day. The CyA regimen was concentration-controlled; that is, subsequent oral doses were selected in an attempt to achieve an average concentration (C_{av}) of 550 ± 50 ng/ml [6]. The clinical protocol for pharmacokinetic monitoring was approved by our institutional Committee for the Protection of Human Subjects. Pharmacokinetic profiles were always performed after administration of at least three equal CyA doses. When C_{av} values were outside the therapeutic range, CyA doses were adjusted in linear fashion to reach the target level. Patients were instructed to ingest their CyA at a fixed time each day. None of the patients in the study was prescribed either drugs known to produce pharmacokinetic interactions with CyA or additional immunosuppressants other than corticosteroids, which were tapered according to the standard schedule previously described [2]. This study only sought to compare CyA pharmacokinetics. None of these 22 subjects experienced a rejection episode during the observation period.

Pharmacokinetic analysis

Aliquots of whole blood (2 ml) were collected just before (0 h), as well as 2, 4, 6, 8, and 12 h after, CyA administration. The samples were withdrawn into tubes containing ethylenediaminetetraacetic acid disodium (EDTA) and analyzed with a specific monoclonal antibody-based fluorescence polarization immunoassay (TDx, Abbott Diagnostic Laboratories, Abbott Park, Ill.). The area under the concentration-time curve (AUC) value of each profile was calculated by the trapezoidal method [10]. In addition, the concentration-time profiles yielded values for the pre-dose (C_0), peak (C_{max}), and final (C_{12}) concentrations, as well as the time to maximum (t_{max}) concentrations, the oral clearance rate (CL/F), and the relative bioavailability [AUC/dose (mg)]. Drug concentrations were analyzed during five post-transplant periods: (1) days 0–2, (2) days 3 and 4, (3) days 5–8, (4) days 9–16, and (5) days 17–30. Patients in the CyA-ME group underwent a median number of eight full pharmacokinetic studies (range 6–10 profiles each), whereas those in the CyA-GC group underwent a median number of seven studies (range 6–14 profiles each; NS, *t*-test; $P = 0.83$).

Statistical methods

A general linear model (GLM) for repeated measures [8] was used to determine the differences between the two treatment groups (CyA-GC vs CyA-ME) during each of the five time intervals with regard to CL/F, t_{max}, fluctuation index {FI; defined as $[(C_{max}/\text{dose})-(C_{min}/\text{dose})]/(C_{av}/\text{dose})$} C_{av}, CyA dose, as well as dose-corrected (meaning divided by dose in mg) C_{av}, C_0, C_{12}, and C_{max} values. The correlations between the AUC and C_0 or C_{12} were evaluated with Pearson's coefficient. For each of the five time intervals, we evaluated the within-group effects and contrasts using simple (time interval) and crossed (time-interval*treatment group) models. A reverse Helmert method [8] was used to examine within-group comparisons, and differences between the two treatment groups were evaluated during each of the five time intervals, as well as for the whole observation period. *P* values less than 0.05 were considered statistically significant. The probability of rejecting a false null hypothesis (power) for this repeated measures design was calculated for the between-group and within-group effects as determined with the simple and the crossed models ($\alpha = 0.05$), respectively. We found that a null hypothesis could be reasonably accepted for the within-group effects (both simple and crossed models; power = 0.83), whereas the null hypothesis was not conclusive for the between-group effects (power = 0.43). The GLM was performed using SPSS software (Version 7.0 for Windows 95, SPSS, Chicago, Ill.) [7]. The power analysis was performed using NCSS/PASS software (Power Analysis and Sample Size, Version 1.0, Dr. Jerry Hintze, Kaysville, Utah). All statistical analyses were performed on an IBM-compatible personal computer with a Pentium processor.

Table 1 Demographic characteristics of patients in the CyA-ME and CyA-GC treatment groups

Demographic characteristic		CyA-ME ($n = 11$)	CyA-GC ($n = 11$)	*P*
Age (years)		41.0 ± 14.11	35.27 ± 9.77	0.014[a]
Gender				NS[b]
	Male	6	8	
	Female	5	3	
Race				NS[b]
	Caucasian	4	9	
	African-American	3	1	
	Hispanic	4	1	
Pretransplant dialysis				NS[b]
	Yes	9	5	
	No	2	6	
Hemodialysis				NS[b]
	Yes	9	7	
	No	2	4	
Peritoneal dialysis				NS[b]
	Yes	1	3	
	No	10	8	
HLA mismatch (A+B+DR)				0.024[a]
	Mean ± SD	4.91 ± 1.14	3.27 ± 1.85	
	0	0	1	
	1–2	0	3	
	3–4	5	4	
	5–6	6	3	

[a] *P* values determined by Student's *t*-test
[b] *P* values determined by Fischer's exact test

Results

Table 1 shows that the two treatment groups were well matched with regard to demographic characteristics, although the mean age of the CyA-ME group was slightly older than that of the CyA-GC group. Table 2 summarizes, whereas Fig. 1 graphically illustrates, the mean

Table 2 Pharmacokinetic parameters of patients treated de novo with either the microemulsion (CyA-ME; $n = 11$) or the gel capsule (CyA-GC; $n = 11$) formulation of cyclosporin (CyA) during serial time intervals immediately post-transplant

	Mean ± SD[a]									
Time	Days 0–2		Days 3–4		Days 5–8		Days 9–16		Days 17–30	
Formulation Parameter[b]	CyA-ME	CyA-GC	CyA-ME	CyA-GC	CyA-ME	CyA-GC	CyA-ME	CyA-GC	CyA-ME	CyA-GC
Dose (mg/kg per 12 h)	5.09 ± 0.83	5.22 ± 0.96	5.60 ± 0.93	7.37 ± 2.00	5.17 ± 1.38	7.83 ± 2.60	4.86 ± 1.42	8.66 ± 6.26	3.62 ± 0.78	6.13 ± 2.94
C_{av} (ng/ml)	265 ± 227	279 ± 149	679 ± 166	486 ± 225	662 ± 218	622 ± 252	700 ± 164	751 ± 266	636 ± 38	781 ± 124
C_{12}(ng/ml)	239 ± 135	191 ± 132	339 ± 166	275 ± 112	339 ± 107	394 ± 191	308 ± 73	375 ± 164	275 ± 62	307 ± 144
C_{max} (ng/ml)	522 ± 358	407 ± 179	1362 ± 362	905 ± 414	1383 ± 553	967 ± 482	1586 ± 457	1428 ± 631	1541 ± 298	1758 ± 735
Dose-corrected C_{av}	0.67 ± 0.46	0.68 ± 0.33	1.49 ± 0.42	0.88 ± 0.43	1.76 ± 0.50	1.07 ± 0.37	2.05 ± 0.43	1.41 ± 0.69	2.43 ± 0.32	1.90 ± 0.67
Dose-corrected C_{12}	0.61 ± 0.29	0.44 ± 0.24	0.73 ± 0.36	0.49 ± 0.22	0.93 ± 0.36	0.68 ± 0.34	0.91 ± 0.23	0.70 ± 0.34	1.01 ± 0.26	0.72 ± 0.36
Dose-corrected C_{max}	1.35 ± 0.81	0.99 ± 0.42	3.02 ± 0.97	1.66 ± 0.81	3.60 ± 1.24	1.67 ± 0.70	4.64 ± 1.21	2.70 ± 1.43	5.84 ± 1.20	4.17 ± 2.08
t_{max}	4.18 ± 2.75	4.55 ± 2.98	3.09 ± 1.38	4.00 ± 2.68	2.36 ± 0.80	4.91 ± 3.15	2.36 ± 0.80	3.09 ± 1.64	2.00 ± 0.00	3.45 ± 1.57
FI	2.38 ± 1.57	0.92 ± 0.48	1.64 ± 0.63	1.35 ± 0.67	1.53 ± 0.56	0.82 ± 0.49	1.78 ± 0.32	1.27 ± 0.37	1.98 ± 0.41	1.79 ± 0.98

[a] Statistical significance of differences between formulations at given time intervals is indicated in the text
[b] For definitions of terms, see Materials and Methods

pharmacokinetic parameters of patients in the two groups. Although the mean C_{av} values were similar for the CyA-ME and CyA-GC groups from days 0 to 2 (265 ± 227 ng/ml and 279 ± 149 ng/ml, respectively; $P = 0.35$), beginning on days 3–4, the mean C_{av} value of the CyA-ME group (679 ± 166 ng/ml) was significantly greater than that of the CyA-GC group (486 ± 225 ng/ml, within-group effect, crossed model, $P = 0.041$; Fig. 1 a). Thus, patients in the CyA-ME group reached target CyA concentrations (C_{av} 550 ± 50 ng/ml) on days 3–4. As the fraction of administered drug that was absorbed continued to increase, the administered dose of CyA-ME was reduced significantly sooner and more rapidly than CyA-GC during the subsequent time intervals ($P = 0.018$; Fig. 1 b). To wit, the patients in the CyA-GC group required significantly higher ($P = 0.023$) and steadily increasing CyA doses through the day 9–16 interval. Thus, the mean dose-corrected C_{av} for the CyA-ME group was significantly higher than that for the CyA-GC group for each of the four time intervals after the day 0–2 period (between-group effect, $P = 0.002$; Fig. 1 c). Although the mean dose-corrected C_{av} values for both treatment groups steadily rose over the course of the entire observation period (within-group effect, $P = 0.026$), the increase in the mean dose-corrected C_{av} for the CyA-ME group was particularly greater than that of the CyA-GC group between the first (days 0–2) and second (days 3–4) time intervals (within-group contrast, crossed model time-intervals*treatment group, $P = 0.001$).

For each of the five time intervals, the mean dose-corrected C_{12} value was significantly higher in the CyA-ME than in the CyA-GC group (Fig. 2a; between-group effect, $P = 0.001$). The mean dose-corrected C_{12} values increased significantly (within-group effect, $P = 0.01$) and proportionately (within-group contrast, crossed model, $P = 0.93$) for both treatment groups. Although the mean dose-corrected C_{max} values for both treatment groups steadily increased (within-group effect, $P = 0.032$) after day 2, they were higher in the CyA-ME than in the CyA-GC group (Fig. 2 b). Moreover, the increase in mean dose-corrected C_{max} was more pronounced in the CyA-ME versus the CyA-GC group from days 0–2 to days 3–4, and from days 3–4 to days 5–8 (within-group contrast, cross model, $P = 0.015$ and $P = 0.02$, respectively).

The mean t_{max} (Fig. 2 c) was significantly shorter in the CyA-ME than in the CyA-GC group during all time intervals (between-group effect, $P = 0.014$) and decreased significantly in both treatment groups over the course of the entire observation period (within-group effect, $P = 0.036$). Although the mean FI was significantly higher in the CyA-ME group (overall between-group contrast, $P = 0.002$; Fig. 2 d), there was no difference after 30 days of therapy (between-group effect, $P = 0.7$). The coefficient of determination (r^2) between the total exposure to CyA (AUC) and the trough level (C_0) for the CyA-ME group ($r^2 = 0.71$) was greater than that for the CyA-GC group ($r^2 = 0.42$; data not shown).

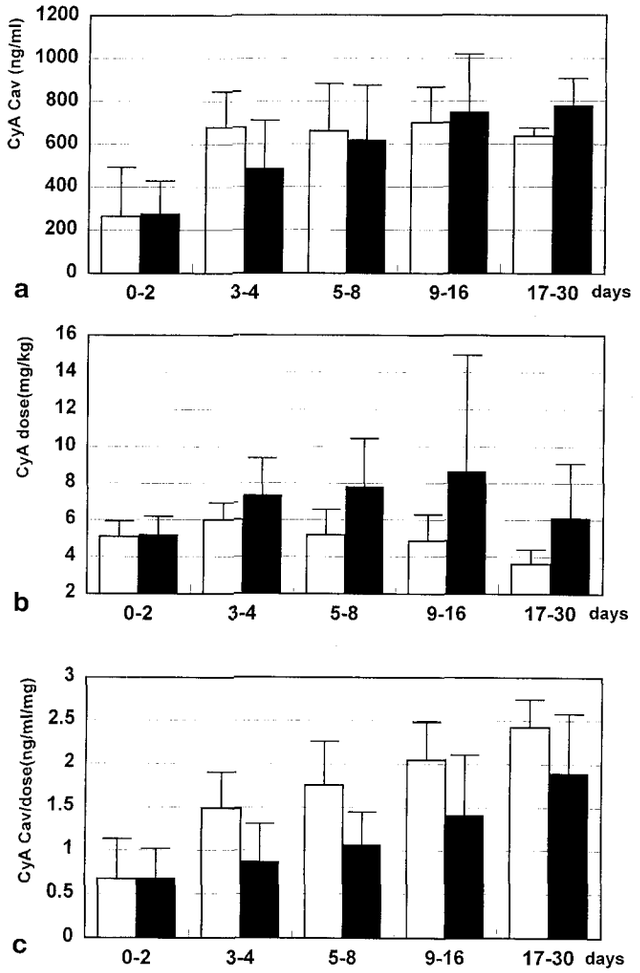

Fig. 1a–c Cyclosporin (CyA): **a** average concentration (C_{av}), **b** dose, and **c** C_{av}/dose over time among patients treated with either the microemulsion formulation of CyA (CyA-ME; □) or the gel capsule formulation of CyA (CyA-GC; ■) expressed as mean values ± standard deviation

Discussion

De novo therapy with oral CyA is complicated by erratic absorption caused by postoperative paralytic ileus as well as by the interindividual pharmacokinetic variability that obscures the relationship between drug dose and exposure [1]. These factors present obstacles to de novo CyA therapy, which demands that the physician establish therapeutic drug concentrations as early as possible in order to achieve a sufficient immunosuppressive effect to avert early allograft rejection [6]. The present study examined whether the previously documented potential advantage of CyA-ME, namely, a reduced dependence on intestinal factors resulting in a greater degree of oral bioavailability, which had been observed for healthy subjects [5] and for stable renal transplant patients [3, 4], also applied in the de novo setting. The present findings confirmed that expectation, and thus suggest that CyA-ME may be used as de novo therapy without an initial umbrella of either intravenous CyA or antilymphocyte antibody induction therapy.

Although neither CyA-ME nor CyA-GC was absorbed well by patients during the first 2 postoperative days, CyA-ME displayed remarkably enhanced absorption by days 3–4. The mean C_{av} after CyA-ME administration was higher than, and reached the target level earlier than, that achieved after CyA-GC administration. In contrast, patients treated with CyA-GC did not achieve target CyA concentrations until days 5–8. The clinical impact of the 48-h delay to establish therapeutic CyA concentrations on the risk of allograft rejection will only be clarified by large trials of de novo CyA-ME therapy.

The present study documents the utility of serial pharmacokinetic studies to assess drug absorption and to adjust drug doses in the de novo setting. The correlation between AUC and trough level ($r^2 = 0.71$) was better for CyA-ME than for CyA-GC ($r^2 = 0.45$). However, a correlation coefficient of 0.7 is not sufficiently reliable to base clinical therapy upon, because it signifies a 30 % error rate among de novo renal transplant patients. Interestingly, although the dose-corrected C_{12} increased proportionately in both treatment groups, only the dose-corrected C_{av} and C_{max} values in the CyA-ME group demonstrated the more pronounced increases from the first to the second time interval, as shown by the significant P value of the crossed model.

At the completion of the study, the drug doses to achieve therapeutic C_{av} levels were 3.62 mg/kg b.i.d. among patients in the CyA-ME group, and 6.13 mg/kg b.i.d among those in the CyA-GC group (t-test; $P = 0.02$). The dose ratio of CyA-ME to CyA-GC is 0.6, the same value as that found by Kovarik et al. in studies of healthy volunteers [5]. However, it is likely that CyA-GC absorption further improves over the first 90 days, so that among stable renal transplant patients the conversion factor is closer to 1.0 [3]. The pharmacokinetic data presented herein demonstrate a substantial advantage of CyA-ME over CyA-GC during de novo therapy to reduce both the time interval and the dose required to achieve target therapeutic concentrations.

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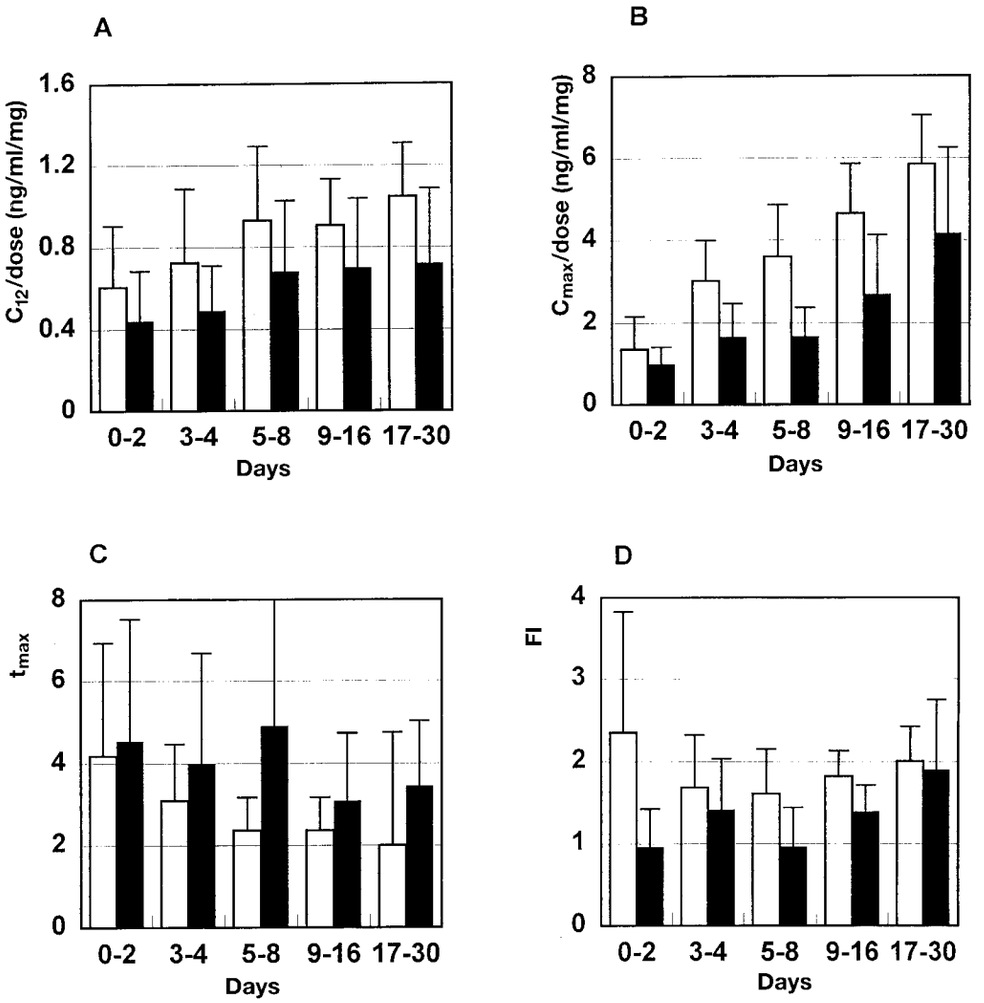

Fig. 2A–D The dose-corrected **A** 12-h (C_{12}) and **B** maximum (C_{max}) CyA concentrations, **C** time to C_{max} (t_{max}), and **D** fluctuation index (FI) among patients treated with either the microemulsion formulation of CyA (CyA-ME; □) or the gel capsule formulation of CyA (CyA-GC; ■) expressed as mean values ± standard deviation

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