

Original Article

Therapeutic Monitoring of Cyclosporine by Determination of C₂ and AUC₀₋₄ Levels in the First Two Years after the Kidney Transplantation

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Abstract

Background. Therapeutic monitoring (TM) of cyclosporine (CyA), by measuring drug concentrations (conc.) in the blood two hours after the administration (C₂), and according to the calculated value of the area under the concentration – time curve in the first four hours after the application (AUC₀₋₄), shows a good correlation with the clinical manifestations in the transplanted patients (pts). The aim of this study was to investigate possibilities of reaching target C₂ and AUC₀₋₄ levels in the early phase after kidney transplantation (Tx), to analyze the administered doses and conc. of CyA in the first two years after Tx and to determine, as well, which of the individual concs. in the early phase after the drug administration shows the best correlation with the AUC₀₋₄.

Methods. In 25 pts (living donor kidney recipients treated by immunosuppressive therapy including Pred, MMF, CyA), the doses of CyA were adjusted after the Tx according to target levels of C₂ and AUC₀₋₄.

Results. The average daily doses of CyA in the first 14 days ranged from 9,6 to 10,1 mg/kg (the largest dose was given on the sixth day). The target C₂/AUC₀₋₄ was reached on the sixth day in 36,3% of pts, on the 9th day in 62,5% of pts. and, on the 14th day in 76% of pts. In each monitored time interval, the target AUC₀₋₄ in relation to C₂, was reached by most of the pts. The maximum CyA concs were the highest 2 hours after the administration (C₂), as compared with the concs after the first and the third hour (C₁ and C₃). In comparison with C₁ and C₃, C₂ showed the best correlation with AUC₀₋₄. 68% of pts displayed the signs of acute CyA nephrotoxicity in the first year and, in 28% of pts the administration of the drug was interrupted in the first two years. A stable graft function was maintained in the first two years after the transplantation.

Conclusion. The highest CyA concentrations in our pts were determined 2 hours after the drug administration, whereas C₂ showed the best correlation with AUC₀₋₄. A considerable number of pts displayed the signs of nephrotoxicity.

Keywords: cyclosporine monitoring, C₂, C₀, immunosuppression, kidney transplantation

Introduction

Although it has already been known that the clinical effect of the CyA depends on the reached conc. levels in the blood, the best way of TM of this drug is not universally

accepted. The measurement of the drug conc. level in the blood prior to the administration of the next successive dose (C₀) and dosing of the drug on the basis of this drug conc., represent the more common way of CyA TM [1]. Recently, it has been shown that C₀ does not correlate well with the episodes of acute rejection, and undesirable drug effects as well [2]. The latest studies shown that the absorption, but not the elimination phase, is primarily significant for the CyA effectiveness [3], and that the area under the concentration-time curve in the first 4 hours after the drug administration (AUC₀₋₄) adequately reflects the exposure of the organism to the drug and correlates well with clinical events [4]. It was also shown that, out of all drug concentration levels separately measured at some points of time during the absorption phase, the drug conc. level determined 2 hours after the administration (C₂) correlates the best with AUC₀₋₄ [5]. Recommended target levels of C₂ and AUC₀₋₄ [6] mostly refer to the time interval of up to one year after Tx, but there are no generally accepted recommendations for desirable conc. levels of those parameters to be applied in the future. However, the significant inter-individual, racial and other specificities impose the need to investigate the feasibility of performing of this type of TM in one's own pts.

Patients and methods

Study group included 25 pts (living donor kidney recipients). The patients who underwent cadaveric kidney transplantation or kidney retransplantation, patients receiving induction therapy with biological antibodies and patients in whom the titer of cytotoxic antibodies in the serum was found to be 50% higher before Tx were excluded. After Tx, the triple immunosuppressive therapy was introduced in all those pts. It included corticosteroids (Methylprednisolon-Lemod Solu, Hemopharm; Pronison, Galenika) administered according to the local department regimen, mikofenolat mofetil (CellCept, RocheLaboratories) in the doses of 1,0 gr every 12 h, administered 2 days before Tx, as well as cyclosporin (Neoral, Novartis Pharma). Cyclosporin was administered perorally and its administration started on the day of Tx, at 05:00 h early in the morning with 6 mg/kg dose. Next morning, when the creatinine level in the blood was 30% lower in relation to the levels measured before Tx, the daily

doses of the drug were increased to 10-12 mg/kg and administered in two daily doses, every 12 h (at 08:00 and 20:00 h).

Adjustment of CyA doses was done according to the algorithm presented by Cole *et al.* [7]. The first determination of CyA conc. was done on the 3rd day after Tx, two hours after administration of the morning drug dose. On the 6th day after Tx, drug conc. was measured immediately before the administration of the morning dose (C0) and continued to be measured after the first (C1), the second (C2) and the 3rd (C3) hour after the drug administration. On the basis of the obtained measures, the AUC level was determined for the first three hours after the drug application (AUC0-3) according to the regressive formula based on C1, C2 and C3 ($AUC_{0-3} = 256 + C1 + 0,9 \times C2 + 1,4 \times C3$) which was proved to have a good correlation ($r=0,99$) with AUC0-4 and AUC0-12 as well⁸. The target C2 level on the 6th day after Tx was 1700 ng/ml, while AUC0-4 level was 4400 ng/ml. The blood sample for C2 was taken 2 hours \pm 15 min. after the administration of the morning drug dose (usually at 10,0h \pm 15 min).

CyA concentrations were measured by fluorescent polarized immunoassay (FPIA, TDx Abbott Laboratories, Chicago, IL, USA) at the Institute of Experimental Medicine and Immunology, Military Medical Academy. The laboratory in which the CyA conc. were determined is a member of the international network for controlling the quality of CyA testing (Bioanalytical Services Ltd, London, UK) and the control is carried out once a month. The mentioned method is a commercial method of the Abbot Company and the detailed instruction may be obtained from a manufacturer.

CyA nephrotoxicity was defined as more than 30% increase in serum creatinin that was not attributed to any other identifiable cause and that improved with decrease in the CyA dose.

Kidney function was measured by serum creatinin levels and calculated creatinine clearance (Cockcroft-Gault).

The patients were informed about the aims and the methods of conducting this research study. Before admitting those pts to the study, they were required to give the written informed consent.

Results

The study group included 16 male (64%) and 9 female pts (36%) aged 17-48 (32,5 on average). Before Tx, 14 pts (56%) underwent hemodialysis and 3 pts (12%) underwent peritoneal dialysis while pre-dialysis Tx was performed in 8 pts (32%). Duration of hemodialysis prior to Tx was 14,1 months on average. Apart from the parents, we had a brother and a sister as donors in 2 cases, relatives of the second generation in another 2 cases and a wife in case of 1 pts. The average age of donors was 54,9 (35-68). The average time of cold ischemia was 27,5 min., of warm ischemia was 78,6 sec. and the time of rewarming was 31,7 min.

The average doses of CyA (mg/kg) applied in the study group of patients in the first 24 months after Tx are presented in the Table 1.

Table 1. The average doses of CyA (mg/kg) applied in the study group of pts. in the first 24 months after Tx

	CyA doses mg/kg / SD
3 days	9,6 / 0,6
6 days	10,1 / 0,8
14 days	9,3 / 1,1
1 months	6,6 / 2,3
3 months	7,2 / 2,8
6 months	4,8 / 1,8
12 months	3,8 / 1,6
24 months	3,7 / 1,5

The average concentrations of cyclosporine (ng/ml) after the first (C1), the second (C2) and the 3rd (C3) hour upon the administration of the drug and, the levels of AUC0-4 measured in the monitored time intervals within 24- month period are presented in the Table 2.

Table 2. The average cyclosporine concs. (ng/ml) after the first (C1), the second (C2) and the 3rd (C3) hour upon the administration of the drug and the levels of AUC0-4 measured in the monitored time intervals within 24- month period

	C1 \pm SD	C2 \pm SD	C3 \pm SD	AUC ₀₋₄ \pm SD
3 days	1108,0	1017,5 \pm 344,3		
6 days	1167,1 \pm 335,9	1342,7 \pm 415,1	1069,7 \pm 315,2	4028,0 \pm 839,6
9 days	1414,0 \pm 330,9	1416,0 \pm 287,2	1053,4 \pm 172,6	4545,2 \pm 497,0
14 days	1518,6 \pm 414,4	1548,0 \pm 316,0	1174,5 \pm 316,0	4798,5 \pm 881,3
1 months	1516,4 \pm 371,4	1733,3 \pm 552,6	1071,7 \pm 282,1	5557,0 \pm 350,4
3 months	1278,5 \pm 471,6	1232,3 \pm 122,7	779,0 \pm 245,2	3744,0 \pm 646,6
6 months	1128,5 \pm 343,4	1075,4 \pm 16,2	723,3 \pm 179,6	3281,3 \pm 579,8
12 months	766,8 \pm 345,3	813,6 \pm 323,7	504,4 \pm 190,3	2514,3 \pm 657,8
24 months	588,0 \pm 221,7	535,7 \pm 207,3	314,0 \pm 89,0	1546,4 \pm 248,7

Table 3. Correlation (r) of AUC0-4 with C1, C2 and, C3 on the 6th and the 9th day after Tx

	AUC ₀₋₄ - C1		AUC ₀₋₄ - C2		AUC ₀₋₄ - C3	
	r	P value	r	P value	r	P value
6. day	0,34	0,011	0,85	0,001	0,58	0,003
9. day	0,38	0,52	0,87	0,001	0,57	0,036

Correlation (r) of AUC0-4 with C1, C2 and, C3 on the 6th and the 9th day after Tx is presented in the Table 3.

One of the target C2/AUC0-4 levels was reached on the 6th day by 36,3% of pts., on the 9th day by 62,5% of pts and, on the 14th day by 76% of pts while, on the 30th day, 84% of patients reached that concentration level. Five pts didn't manage to reach the target conc. levels in the first 30 days after Tx. By separate analysis of C2 and AUC0-4, it could be observed that the target C2 conc. level was reached on the 6th day by 6 pts (27,2%) and the target AUC0-4 level

by 7 pts (31,8%, $P=0,95$). The target C2 was reached by another 2 pts (33,3%) on the 9th day and the target AUC0-4 level was reached by another 6 pts (54,1%, $P=0,35$). On the 14th day after Tx, 11 pts (44%) reached the target C2 level and 17 pts. (68%, $P=0,23$) reached the target AUC0-4 level. The signs of cyclosporine nephrotoxicity were displayed by 44% of pts in the first 14 days, and by 68% of pts in the first year. Administration of CyA in the first two years was permanently interrupted in 7 pts (28%) due to the impaired graft function and the possible impact of the cyclosporine nephrotoxicity. The average creatinine clearance in the study group was 65 ml/min after 3 months, 66,2 ml/min after 6 months, 61,5 ml/min after 12 months and 60,8 ml/min, after 24 months. There was no loss of graft reported within the monitoring period.

Discussion

Recent studies shown that the desirable CyA efficacy may be achieved if adequate drug concentration levels in the blood are reached in the initial phase, up to the 7th day after the Tx [8].

The average CyA doses used in our study were very similar to the doses applied in other studies in which TM was performed by C2 monitoring: as presented in a comprehensive, randomized and multicenter study (MO2ART) [9], the average daily dose of the drug on the 5th day after Tx was 10,3 mg/kg, and 11 mg/kg in the Canadian study [10]. The drug dose started to be reduced after Tx and, after one month, it was 35% (6,6±2,3) smaller in relation to the initial dose. The rapid reduction of the drug dose was also described in the MO2ART study [9], according to which, the average daily dose of CyA after one month was 6,7 mg/kg.

Opelz *et al* investigated the relationship between the daily doses of CyA one year after Tx with the long-term kidney graft function in the group of cadaveric kidney recipients [11]. The results indicated that the 7-year graft survival period in pts receiving CyA after the first year in the doses below 2 mg/kg was the shortest, whereas, it was the longest in the group of pts receiving the average daily doses of about 5 mg/kg. However, the graft survival was also very short in pts receiving daily doses of over 6 mg/kg of CyA. The average daily CyA dose in the group of our pts one year after Tx reached the medium of the mentioned levels (3,8±1,6) and the administered drug doses were relatively constant till the second year (3,7±1,5) We think that maintenance of the stable CyA conc levels (even within the first two years after Tx) may be very important for the long-term graft function.

In our pts in the first two weeks after Tx (except for the 3rd day), the maximum CyA conc. level was determined 2 hours after the drug administration, while C2 shown the best correlation with AUC0-4, what was confirmed in other studies [12,13]. Such findings could justify the choice of C2 as the most appropriate separate point for TM of CyA in our patients.

Apart from relatively high drug doses given immediately after Tx, only more than one third of our patients managed to reach the target C2 or AUC0-4 conc. levels on the 6th day, what constituted to be a problem observed in other studies in which TM was managed by C2. In the Italian study [14], the target C2 of 1700 ng/ml was reached by

50% of patients on the 7th day and, according to the Canadian study¹⁵, the target AUC0-4 conc. level was reached by 49% of pts on the same day. Taking into consideration the fact that the target AUC0-4 levels in relation with C2 levels in each monitored time intervals were reached by a larger number of our pts, it may be observed that, in nearly 25% of our pts, the CyA dose would be increased in the first 14 days after Tx if the drug TM is managed by C2 level monitoring only.

Conclusion

In the group of pts in which TM was performed by C2/AUC0-4 level monitoring, the highest conc. of CyA was determined in C2, which correlated well with AUC0-4. Since the smaller number of pts reached the target C2/AUC0-4 level in the early monitoring period and that a considerable number of pts displayed the signs of nephrotoxicity, the target C2/AUC levels could be lower than that of recommended for our pts. The graft function was stable in the study group.

Conflict of interest statement. None declared.

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