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Cyclosporine Metabolite Profiles in the Blood of Liver Transplant Patients

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CYCLOSPORINE (CyA) is a potent immunosuppressive agent that undergoes extensive hepatic metabolism. Eleven metabolites of CyA have been fully characterized.[1] Recent reports indicate that CyA metabolites have in vitro immunosuppressive activity and/or nephrotoxicity.[2-4] The time course of the formation and elimination of these metabolites in the blood of transplant patients has not been documented, however. The objective of our study, therefore, was to quantitate four major CyA metabolites in the blood of liver transplant patients over a dosing interval after oral and intravenous CyA administration.

MATERIALS AND METHODS

Patients

Three adult liver transplant patients who were clinically stable, on the basis of serum bilirubin and liver enzyme levels, were enrolled in the study. Blood sampling was conducted on two consecutive days: after the patient's oral CyA dose on the first day and after an intravenous CyA dose that was 50% of the oral dose on the second day. Hourly blood samples were obtained during the 12-hour dosing interval on both study days.

Measurement of CyA and Metabolites

The measurement of CyA and metabolite concentrations used a gradient high-performance liquid chromatographic (HPLC) system. The column was a Resolve C-18, 15 cm × 3.9 mm, 5-μm column (Waters, Milford, MA) that was heated to 70°C. The mobile phase consisted of a linear gradient of acetonitrile and water delivered at 1.0 mL/min. Two milliliters of whole blood was mixed with an internal standard (cyclosporine D) and extracted with diethyl ether. The final dried extract was reconstituted in 100 μL methanol, and 40 μL was injected

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Table 1. Blood Concentrations of CyA and Metabolites in Liver Transplant Patients

Route (Dose, mg/kg)	Average Concentration* (ng/mL) Range				
	CyA	M17	M1	M18	M21
Patient 1					
IV (2.9)	1,253	1,200	369	161	161
	(203-3,271)	(441-1,570)	(109-485)	(57-223)	(53-286)
PO (5.7)	808	1,301	544	213	202
	(356-1,578)	(550-1,860)	(117-898)	(38-349)	(41-364)
Patient 2					
IV (1.7)	593	860	226	91	72
	(203-1,537)	(601-1,358)	(115-297)	(44-186)	(32-136)
PO (3.5)	364	648	238	86	46
	(232-571)	(535-1,349)	(125-480)	(56-203)	(41-145)
Patient 3					
IV (2.2)	695	647	279	53	66
	(170-2,306)	(434-860)	(153-412)	(<25-86)	(<25-141)
PO (4.4)	393	464	280	24	52
	(235-635)	(344-660)	(81-542)	(<25-59)	(<25-183)

Abbreviations: IV, intravenous; PO, oral.

*Average steady-state concentration (Cp^{ss}) calculated as area under the curve divided by the dosing interval.

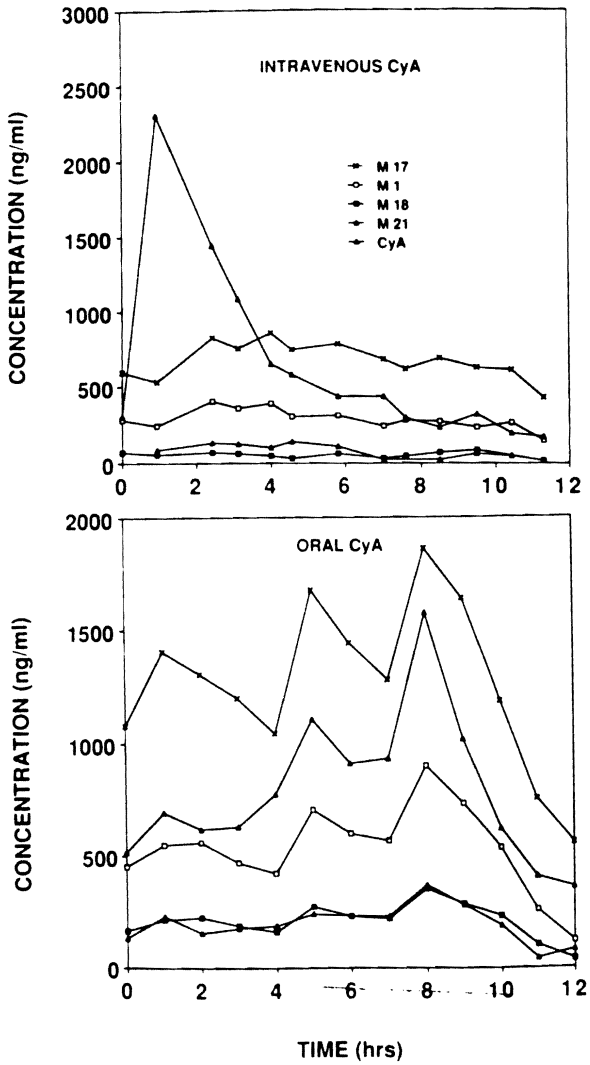

Fig 1. CyA and metabolite blood concentrations after liver transplantation.

onto the HPLC column. Standards of the metabolites (M17, M1, M18, M21) were prepared from material recovered from human bile and confirmed by HPLC and mass spectrometry. Standard curves were prepared for CyA and each of the four metabolites that were quantified.

RESULTS

The average and range of blood concentrations observed during the dosing interval are presented in Table 1. Figure 1 demonstrates the typical metabolite concentrations measured over time during the dosing interval. The blood concentrations for the three patients were M17 > CyA > M1 for the oral study, whereas M18 and M21 concentrations varied during the dosing interval. In the intravenous study, the blood concentrations were CyA > M17 > M1 in two patients, with the third patient following the oral pattern. Metabolite concentrations did not consistently decline over the dosing interval; the CyA and metabolite concentrations increased to their highest level between nine and 12 hours in the oral study of patient no. 2.

DISCUSSION

High concentrations of CyA metabolites, particularly M17 and M1, were present in the

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7 and M1, were present in the blood of liver transplant patients. Concentrations of and total exposure to M17 exceed or approximate those of CyA in these patients. Because both M17 and M1 are immunologically active,[2] a contribution to the immunosuppressive effect of CyA in liver transplant patients is possible. During the dosing interval, extreme variability in metabolite blood concentrations was observed. The late increase in metabolite concentrations in the oral dosing interval of patient 2 may be due to either altered CyA absorption or metabolism. Additional studies are necessary to evaluate the immunosuppressive or toxic effects of these high concentrations of blood metabolites in liver transplant patients.

REFERENCES

1. Maurer G, Lemaire M: Transplant Proc 18:25, 1986 (suppl 5)
2. Freed BM, Rosano TG, Lempert N: Transplantation 43:123, 1987
3. Yee GC, Kennedy MS, Self SG, et al: Transplant Proc 18:774, 1986
4. Zeevi A, Venkataramanan R, Burckart G, et al: Hum Immunol (in press)