

Original Article

Safety of reduced dose of mycophenolate mofetil combined with tacrolimus in living-donor liver transplantation

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Background/Aims: The dose of mycophenolate mofetil (MMF) has been reduced in Asia due to side effects associated with the conventional fixed dose of 2-3 g/day. We aimed to determine the pharmacokinetics of a reduced dose of MMF and to validate its feasibility in combination with tacrolimus in living-donor liver transplantation (LDLT).

Methods: Two sequential studies were performed in adult LDLT between October 2009 and 2011. First, we performed a prospective pharmacokinetic study in 15 recipients. We measured the area under the curve from 0 to 12 hours (AUC₀₋₁₂) for mycophenolic acid at postoperative days 7 and 14, and we performed a protocol biopsy before discharge. Second, among 215 recipients, we reviewed 74 patients who were initially administered a reduced dose of MMF (1.0 g/day) with tacrolimus (trough, 8-12 ng/mL during the first month, and 5-8 ng/mL thereafter), with a 1-year follow-up. We performed protocol biopsies at 2 weeks and 1 year post-LDLT.

Results: In the first part of study, AUC₀₋₁₂ was less than 30 mg·h/L in 93.3% of cases. In the second, validating study, 41.9% of the recipients needed dose reduction or cessation due to side effects within the first year after LDLT. At 12 months post-LDLT, 17.6% of the recipients were administered a lower dose of MMF (0.5 g/day), and 16.2% needed permanent cessation due to side effects. The 1- and 12-month rejection-free survival rates were 98.6% and 97.3%, respectively.

Conclusions: A reduced dose of MMF was associated with low blood levels compared to the existing recommended therapeutic range. However, reducing the dose of MMF combined with a low level of tacrolimus was feasible clinically, with an excellent short-term outcome in LDLT. (*Clin Mol Hepatol* 2014;20:291-299)

Keywords: Area under the curve; Mycophenolate mofetil; Mycophenolic acid; Therapeutic drug monitoring; Liver transplant

INTRODUCTION

Mycophenolate mofetil (MMF) has been combined with calcineurin inhibitors for the prevention of acute cellular rejection (ACR) and to decrease the side effects of calcineurin inhibitors following

kidney, heart, and liver transplantation (LT).¹ Despite the widespread use of MMF in combination with tacrolimus, the optimal dose for this combination has yet to be defined, especially for LT.² It has conventionally been administered as a fixed dose (2.0-3.0 g/day) without routine plasma concentration monitoring of myco-

Abbreviations:

ACR, acute cellular rejection; Alc., alcoholic; AUC₀₋₁₂, area under the curve from 0 to 12 hours; BUN, blood urea nitrogen; BWT, body weight; C₀MPA, plasma concentration of mycophenolic acid at pre-dosing; GFR, glomerular filtration rate; GI, gastrointestinal; HBV, hepatitis B virus; HCC, hepatocellular carcinoma; LC, liver cirrhosis; LDLT, living donor liver transplantation; LFT, liver function tests; LT, liver transplantation; MELD, model for end-stage liver disease; MMF, Mycophenolate mofetil; MPA, mycophenolic acid; POD, postoperative days; RAI, rejection activity index; TDM, therapeutic drug monitoring; WBC, white blood cell

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phenolic acid (MPA), the active metabolite.²⁻⁶

In a 2008 consensus meeting about MPA in Rome,⁷ the recommended initial daily dose of MMF was 3.0 g rather than 2.0 g with cyclosporine in adult transplantations. It is more likely to achieve the minimum target MPA exposure in early post transplantation. However, clinically, the most popular dose of MMF is based on the side effects (leukopenia, gastrointestinal problems such as abdominal discomfort or diarrhea, infection, etc.). Indeed, many studies conducted in Asian renal transplant recipients, including a randomized controlled trial, suggested the need for MMF dose reduction so as to minimize the side effects of MMF.⁸⁻¹⁰ In Korea, mostly 1.0 to 1.5 g daily of MMF was used in actual clinical situations, based on the side effect. That is a generally reduced dose in comparison to that given in Western countries and in comparison to the recommendation from the 2008 consensus meeting.

There are a few studies on the pharmacokinetics or feasibility of a reduced dose of MMF, especially in living donor LT (LDLT). Therefore, we designed two sequential studies with the following aims: 1) a preceding prospective preliminary study for pharmacokinetics of a reduced dose of MMF in early post-LDLT, and 2) a validating study for the feasibility of a reduced dose of MMF with tacrolimus combination within the first year follow-up after LDLT.

PATIENTS AND METHODS

Patient selection

For the pharmacokinetics of a reduced dose of MMF in early post-LDLT, we performed a preceding prospective preliminary study, and then a retrospective following study for validation. In the preliminary study, eligible patients were adult LDLT recipients with end-stage liver disease at our institution, and we administered MMF (mainly 1.0 g/day) with tacrolimus (through level, 8-12 ng/mL) with 14 days follow-up after LT (or hospital stay if the length of stay was <14 days). We performed the protocol biopsy before discharge. We excluded patients with any of the followings: age <19 or >65 years, previous organ transplantation, multiple organ transplantation, pregnancy, breast-feeding, advanced hepatocellular carcinoma with invasion of major vessels or distant metastasis, human immunodeficiency virus positive, active peptic ulcer within two weeks, administration of other study drug, hemodialysis within 30 days of LT, serum creatinine level over 1.4 mg/dL before LT, contraindication of tacrolimus or corticosteroids or MMF or basiliximab, and poor compliance to regular follow-up.

This study was approved by the institutional review board at Seoul National University Hospital (approval number: 0811-068-263).

Between October 2009 and December 2011, 365 cases of LT had been performed at the Seoul National University Hospital (Seoul, Korea), and all of recipients received blood group compatible grafts and orthotopic LT. Among these, 215 recipients were adults, and underwent LDLT. In the following validating study, we selected the patients who were initially administered a reduced dose of MMF (1.0 g/day) with tacrolimus, with a follow-up period of 1 year after LT. We excluded patients with same criteria in the preceding pharmacokinetic study and the patients enrolled in the preceding study. This study was approved by the institutional review board at Seoul National University Hospital (approval number: 1311-086-536).

Immunosuppressive protocol

All recipients had received induction with 20 mg of intravenous basiliximab (an interleukin-2 receptor antagonist; Simulect, Novartis) within two hours before LT and on postoperative days (POD) 4. Initially, a basal immunosuppressive regimen based on tacrolimus (Prograf, Astellas or Tacrobell, Chong Kun Dang), steroids, and MMF (CellCept, Roche) was administered as a part of the initial triple immunosuppressive regimen or was introduced during follow-up as a maintenance immunosuppressive agent.

Tacrolimus started within 5 days after LT. The tacrolimus dose was adjusted according to individual clinical need with respective target whole-blood trough levels around 8 to 12 ng/mL for the first month after LT, followed by 5 to 8 ng/mL thereafter. Intravenous methylprednisolone 500 mg was given intra-operatively before portal perfusion, and was tapered from 200 mg to 20 mg within 6 days. Thereafter, oral prednisolone was continued at 20 mg daily until the first month after LT, and tapered to 0 to 5 mg/day after about 6 months post-LT. MMF was started with 1.0 g daily within 5 days (93.3% in the preliminary study and 100.0% in the validating study) after LT and adjusted according to the occurrence of related side effects.

Measurements of MPA plasma concentration and the AUC₀₋₁₂

In the pharmacokinetic study, for therapeutic drug monitoring (TDM) of MMF, we measured the concentration of MPA and the area under the curve from 0 to 12 hours (AUC₀₋₁₂). To measure pharmacokinetic parameters, blood samples before the dose (C₀)

and at 1, 2, 4, 6, and 12 hours after the dose were obtained on POD 7 and 14. About 5 mL of venous blood were collected into EDTA tubes. The plasma concentration of MPA was measured by a validated high-performance liquid chromatography tandem mass spectrometry. The full AUC₀₋₁₂ was determined according to the Classic linear trapezoidal method.

Follow-up and investigation of safety and efficacy

In the validating study, the patients were usually discharged in POD 14 to 30, and complete laboratory investigation was conducted almost daily during hospital stay, including liver function tests (LFT) and blood tacrolimus trough level. Outpatient follow-up was usually conducted once a week for the first month after discharge and was gradually lengthened to every 3 or 4 months, with additional visits as Clinically necessary. A complete laboratory investigation, including LFT and blood tacrolimus trough level measurement, was conducted at each follow-up.

MMF was initially administered daily with dose of 1.0 g if the white blood cell (WBC) count exceeded 3,500/mm³, reduced to 0.5 g daily for 2,500 to 3,500/mm³, and ceased for less than 2,500/

mm³. During follow-up, we decreased or ceased the MMF based on apparent toxicities with clinical suspicion related to MMF: neutropenia (defined as WBC count <3,500/mm³ in this study), gastrointestinal problem, or infection including cytomegalovirus.

Nephrotoxicity was defined as an elevated serum creatinine (>1.4 mg/dL) after excluding renal insufficiency due to other causes.

Patients underwent an ultrasonography-guided protocol liver biopsy. Protocol biopsies were performed at POD 7 to 14, additional biopsies at 1 year after LT, and at the time as clinically indicated. We defined the acute cellular rejection (ACR) when the rejection activity index (RAI, Banff score; 0-2, no rejection; 3, borderline; 4-5, mild rejection; 6-7, moderated rejection; 8-9, severe rejection) was of 4 or more points.¹¹ When RAI was 4 points, we maintained the level of tacrolimus in cases without abnormal LFT, but we increased the level of tacrolimus with significant abnormal LFT results. When RAI was 5 or more points, we treated the patients with steroid pulse therapy regardless of abnormal LFT. In all cases, informed consent was obtained from each patient before biopsy.

Table 1. Characteristics of the patients and pharmacokinetic parameters of a reduced dose of MMF in preliminary study

	Baseline characteristics					On POD 7			On POD 14		
	Age (years)	Gender	BWT (Kg)	Height (cm)	Diagnosis	Dose (g/day)	C ₀ MPA (mg/L)	AUC ₀₋₁₂ (mg h/L)	Dose (g/day)	C ₀ MPA (mg/L)	AUC ₀₋₁₂ (mg h/L)
1	44	Female	77.0	161.0	HBV-LC, HCC	0.00*	-	-	0.50	0.2	14.10
2	55	Male	54.0	170.9	HBV-LC	1.00	1.9	21.50	1.00	1.6	31.15
3	62	Male	64.1	169.5	HBV-LC	1.00	0.2	17.80	1.00	0.2	10.65
4	58	Male	70.5	168.4	HBV-LC	1.00	0.3	6.90	1.00	0.2	12.40
5	48	Male	62.7	174.7	HBV-LC, HCC	1.00	0.8	19.95	1.00	0.1	11.50
6	52	Male	69.2	166.3	HBV-LC	1.00	0.1	5.55	1.00	0.1	4.40
7	56	Male	77.4	173.9	HBV-LC	1.00	0.5	13.90	1.00	1.0	10.30
8	60	Male	42.9	150.8	Biliary-LC	1.00	0.4	15.95	1.00	0.6	9.95
9	51	Male	66.7	170.7	HBV-LC, HCC	1.00	0.4	26.45	1.00	0.4	16.05
10	53	Male	71.6	169.3	HBV-LC, HCC	1.00	0.2	19.65	1.00	0.1	15.65
11	64	Male	55.9	157.0	Alc.-LC, HCC	1.00	0.6	14.25	1.00	†	8.40
12	51	Male	53.7	169.5	HBV-LC, HCC	0.75	1.0	12.25	0.50	0.5	4.20
13	34	Male	54.0	171.0	HBV-LC, HCC	1.00	0.5	19.85	1.00	0.3	8.40
14	37	Male	54.6	173.4	HBV-LC	1.00	0.5	17.30	1.00	0.5	15.65
15	57	Male	50.2	172.0	HBV-LC	1.00	1.2	16.30	1.00	1.0	13.85

MMF, mycophenolate mofetil; POD, postoperative days; BWT, body weight; C₀MPA, plasma concentration of mycophenolic acid at pre-dosing; AUC₀₋₁₂, the area under the curve from 0 to 12 hours; HBV, hepatitis B virus; LC, liver cirrhosis; HCC, hepatocellular carcinoma; Alc., alcoholic.

*Skip the MMF due to pre-dosing neutropenia.

†Not checked.

Table 2. Baseline characteristics of the patients in a reduced dose of MMF with tacrolimus combination in adult LDLT

Factor	Total (n=74)
Gender (male:female)	61:13 (82.4%:17.6%)
Age at LT (years)	52.28±6.37 (range, 25-65)
Diabetes	15 (20.3%)
Hypertension	10 (13.5%)
Primary liver diseases	
Hepatitis B related liver cirrhosis	56 (75.7%)
Hepatitis C related liver cirrhosis	1 (1.4%)
Alcoholic liver cirrhosis	11 (14.9%)
Combined hepatocellular carcinoma	45 (60.8%)
Child-Turcott-Pugh score	8.09±2.38 (range, 5-13)
MELD score	15.32±8.23 (range, 6-44)
Body mass index (kg/m ²)	23.75±3.10 (range, 15.2-32.5)
Donor age (years)	28.93±10.83 (range, 16-58)
Graft-versus-recipient body weight ratio (%)	1.21±0.25 (range, 0.7-1.9)
Preoperative laboratory	
Serum creatinine	0.83±0.22 (range, 0.2-1.4)
BUN	14.59±7.43 (range, 0.2-43)
GFR	104.23±38.50 (range, 53.0-335.5)
Elective: emergency operation	72:2 (97.3%:2.7%)
Operative time (min)	396.49±79.10 (range, 225-625)
Ischemic time (min)	84.78±27.40 (range, 31-165)
Cold ischemic time (min)	50.19±23.26 (range, 7-134)
Warm ischemic time (min)	34.75±12.68 (range, 15-100)
Postoperative hospital stay (day)	14.42±6.69 (range, 9-62)

Data are presented as the mean±SD.

MMF, mycophenolate mofetil; LDLT, living donor liver transplantation; MELD, model for end-stage liver disease; BUN, blood urea nitrogen; GFR, glomerular filtration rate (using Modification of Diet in Renal Disease formula).

Table 3. Side effects of a reduced dose of MMF

Side effects	Overall, n (%)		First month, n (%)		Month 2-6, n (%)		Month 7-12, n (%)	
	Cases (n=74)	↓ or cessation of MMF*	Cases (n=74)	↓ or cessation of MMF*	Cases (n=74)	↓ or cessation of MMF*	Cases (n=74)	↓ or cessation of MMF*
Leukopenia	38 (51.4)	27 (36.5)	15 (20.3)	7 (9.5)	28 (37.8)	20 (27.0)	26 (35.1)	17 (23.0)
GI problem [†]	5 (6.8)	5 (6.8)	0 (0)	0 (0)	3 (4.1)	3 (100)	2 (2.7)	2/2 (100)
Nephrotoxicity	8 (10.8)	0 (0)	1 (1.4)	0 (0)	8 (10.8)	0 (0)	5 (6.8)	0/5 (0)
Infection [‡]	4 (5.4)	4 (5.4)	1 (1.4)	0 (0)	1 (1.4)	1 (100)	2 (2.7)	2/2 (100)
Total (n=74)		31 (41.9)		7 (9.5)		23 (31.1)		26 (35.1)

MMF, mycophenolate mofetil; GI, gastrointestinal.

*Dose reduction or cessation of MMF due to related side effects.

[†]One case accompanying with leukopenia.

[‡]All cases accompanying with leukopenia.

Statistical analysis

In the validating study, statistical analyses were performed using SPSS version 19.0 (SPSS Inc., Illinois, USA). Continuous data were presented as the mean \pm standard deviation or medians, and categorical parameters were presented as the number of cases with percentage in parenthesis. Survival rates were estimated using the Kaplan-Meier method and compared using the Log rank test.

RESULTS

Patient enrollment and characteristics

Among 215 adult patients who underwent LDLT between October 2009 and December 2011, 17 patients were enrolled in the pharmacokinetic study. Two patients dropped out due to withdrawal (refusal of samplings). Finally, 15 patients (14 males and 1 female; mean age 52.1 years, range 34 to 64 years) were enrolled.

LT had been performed mainly due to hepatitis B virus-related liver cirrhosis in 86.7% (n=13), and 7 patients (46.7%) had a coexisting hepatocellular carcinoma. The baseline characteristics of the 15 patients are shown in Table 1.

In the validating study, among 215 adult patients, 74 patients (61 males and 13 females; mean age 52.3 years, range, 25-65 years) were included. LT had been performed due to mainly hepatitis B virus-related liver cirrhosis (75.7%), and 60.8% had a coexisting hepatocellular carcinoma (n=45). Table 2 summarized the baseline characteristics of the 74 patients.

Plasma MPA levels and AUC₀₋₁₂ of reduced dose of MMF (mainly 1.0 g daily)

In the pharmacokinetic study, the initial dose of MMF started with 1.0 g daily with tacrolimus within 5 days after LT and was maintained with 1.0 g daily during follow-up, except for 2 cases: one for pre-dosing leukopenia and the other for side effect. After all, most of the patients received MMF with 1.0 g daily (86.7%) in

Table 4. Short-term outcome of a reduced dose of MMF with low level of tacrolimus in adult LDLT

Outcome	Overall (n=74)	First month (n=74)	Month 2-12 (n=74)
Pathologic outcomes			
RAI score			
0-2 (no ACR)	68/74 (91.9%)	65/69 (94.2%)	59/61 (96.7%)
3 (borderline ACR)	4/74 (5.4%)	3/69 (4.3%)	1/61 (1.6%)
4-5 (mild ACR)	2/74 (2.7%)	1/69 (1.4%)	1/61 (1.6%)
≥ 6 (\geq moderate ACR)	0/74 (0.0%)	0/69 (0.0%)	0/61 (0%)
Biopsy-proven ACR	2/74 (2.7%)	1/69 (1.4%)	1/61 (1.6%)
Portal inflammation	29/74 (39.2%)	13/69 (18.8%)	23/61 (37.7%)
Fibrosis	14/74 (18.9%)	0/69 (0.0%)	14/61 (23.0%)
Rejection-free survival	72 (97.3%)	73 (98.6%)	73 (98.6%)
Major morbidity			
Biliary problem	8 (10.8%)	2 (2.7%)	6 (8.1%)
Abdominal fluid collection	3 (4.1%)	1 (1.4%)	2 (2.7%)
Bleeding	2 (2.7%)	2 (2.7%)	0 (0%)
Hepatic artery stenosis	2 (2.7%)	2 (2.7%)	0 (0%)
Wound repair	1 (1.4%)	0 (0%)	1 (1.4%)
Spleno-renal shunt ligation	1 (1.4%)	1 (1.4%)	0 (0%)
Mortality			
Graft survival	74 (100%)	74 (100%)	74 (100%)
Patient survival	74 (100%)	74 (100%)	74 (100%)

MMF, mycophenolate mofetil; LDLT, living donor liver transplantation; RAI, rejection activity index; ACR, acute cellular rejection.

early post-LDLT. In the first case, the patient had to skip the MMF for a week due to pre-dosing leukopenia and then started with a lower dose (0.5 g/day). In case number 12, the patient's initial dose of MMF was 1.0 g daily, but he had to reduce the dose as time passed because of gastrointestinal symptoms. Comparing to the generally recommended therapeutic range (30-60 mg h/L),⁷ the AUC₀₋₁₂<30 mg h/L was present in 100% on POD 7 (range, POD 6-8) and present in 93.3% on POD 14 (range, POD 10-19). The median AUC₀₋₁₂ was 16.30 mg h/L (range, 5.55-26.45 mg h/L) on POD7 and 11.50 mg h/L (range, 4.20-31.15 mg h/L) on POD14 as shown in Table 1.

Efficacy and safety

In the preliminary study, the mean hospital stay was 15.6 days (range, 9-43 days). There was no mortality and no major complications (Clavien-Dindo classification grade \geq IIIa).¹² There was no biopsy-proven ACR or graft loss. Also, there were no episodes of elevation of blood urea nitrogen or serum creatinine (>1.4 mg/dL). There was only one case of a drug related side effect, case number 12, as previously described. The patient's initial dose of MMF was 1.0 g daily, but he had to reduce the dose as time passed because of gastrointestinal problem. He received the dose of 0.75 g/day on POD 7, and 0.5 g/day on POD 14 (Table 1).

In the validating study, side effects of the patients in immunosuppression of reduced dose of MMF combined with a low level of tacrolimus in adult LDLT are summarized in Table 3. A total of 64.9% (n=48) maintained the initial dose of MMF (1.0 g/day). Among 74 recipients, 41.9% (n=31) experienced dose reduction or cessation of MMF due to related side effects at least once within the first year after LT. Finally, 12 months after LT, 17.6% (n=13) were administered a lower dose of MMF (0.5 g/day), and 16.2% (n=12) needed permanent cessation of MMF due to side effects. Even with a reduced dose of MMF with tacrolimus combination immunosuppression, leukopenia was present in 51.4%, nephrotoxicity in 10.8%, gastrointestinal problems in 6.8%, and infection in 5.4% during the study period. Leukopenia was the most common side effect requiring dose reduction or cessation of MMF. At 1 year after LT, nephrotoxicity related with tacrolimus was present in 4.1% of the cases (n=3), but there was no severe renal impairment requiring medication or renal replacement therapy. One patient needed an increased dose of MMF (1.5 g/day) to reduce tacrolimus due to calcineurin inhibitor related nephrotoxicity.

In the validating study, the short-term outcome of the patients in immunosuppression of a reduced dose of MMF combined with

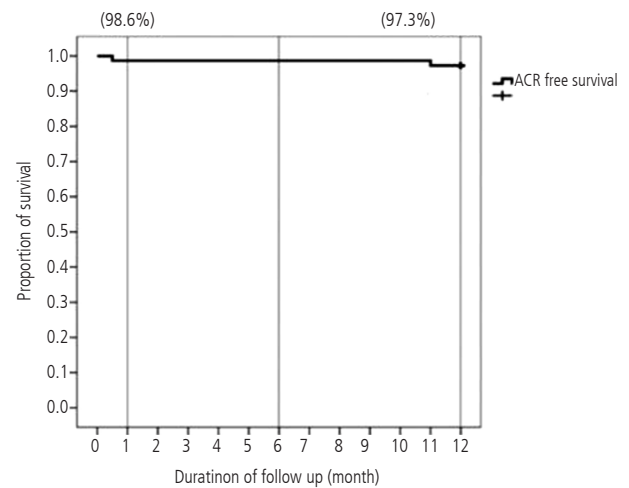


Figure 1. Kaplan-Meier curve showing acute cellular rejection (ACR)-free survival for patients in reduced dose of mycophenolate mofetil with tacrolimus combination in adult living donor liver transplantation.

a low level of tacrolimus with a follow-up of 1 year in adult LDLT is summarized in Table 4. Overall biopsy-proven ACR was present in 2.7% of the cases (n=2), and both cases were of mild ACR. One patient who underwent biopsy at 2 weeks post-LT showed 4 points of RAI without an abnormal LFT and maintained the usual immunosuppressants. The other patient who underwent biopsy at 1 year post-LT showed 5 points of RAI with a significant abnormal LFT and was treated with steroid pulse therapy with good response. In other pathologic abnormalities, portal inflammation (\geq mild degree) was present in 39.2%, and fibrosis (\geq mild degree) in 18.9% of the cases within the first year after LT. Major morbidity (Clavien-Dindo classification grade \geq IIIa) was present in 18.9% (n=14), and biliary complication was most common. There was no mortality. Patient and graft survival were 100% at 12 months after LT, both. The 1-, 6- and 12-month rejection-free survival rates were 98.6%, 98.6% and 97.3%, respectively (Fig. 1).

DISCUSSION

The initial dose of MMF was recommended at 2.0 to 3.0 g daily in previous studies, but those were based on adult renal transplantation and concomitant cyclosporine therapy.^{2-5,7,8} The recommended initial dose of MMF is based on data from clinical trials carried out in Western countries,^{7,8,13} and, actually, lower doses of MMF have been needed due to side effects in clinical settings in Asian countries. Therefore, we designed the present study composed of pharmacokinetics and validation on the appropriate dose

of MMF combined with tacrolimus in LDLT in Asian. To the best of our knowledge, this was the first biopsy-proven study on that.

The MPA AUC_{0-12} is generally regarded as the most reliable pharmacokinetic parameter of risk for ACR^{14,15} and the choice for drug exposure evaluation.¹⁶⁻¹⁸ Although the results are conflicting, MPA levels determined via high performance liquid chromatography between 1.0 and 3.5 mg/L for the C_0 , and 30 to 60 mg h/L for AUC_{0-12} have been recommended following renal or heart transplantation.^{3,4,6,7,14,19,20}

Van Gelder et al.¹⁷ concluded that under-exposure to MPA in the first week after renal transplantation is highly predictive of the development of subsequent acute rejections, and an increased MMF starting dose may be the way to go. However, Brunet et al. reported²¹ that during the first month after LT, exposure to MPA was low in a considerable proportion of 15 LT recipients receiving MMF at 2 g/day and tacrolimus, and they showed that there was no relationship between MPA pharmacokinetics and the efficacy of MMF. In the present study, compared to the generally recommended therapeutic range, the AUC_{0-12} was low in almost all patients except one. However, there was no biopsy-proven ACR, graft loss, or death in early post-LDLT.

From other studies, the incidence of dose reduction of MMF at least once due to side effects ranged from 42.0% to 70.3% with a standard dose of MMF of 2.0 or 3.0 g/day in renal transplantation.²²⁻²⁴ In the present study, although MMF was initially administered in a low dose (1.0 g/day) compared to recommendations from others, 41.9% experienced a dose reduction or cessation of MMF due to related side effects at least once within the first year after LT. Finally, about one-third of patients (33.8%) needed to reduce the dose of MMF (<1.0 g/day) due to related side effects at 1 year post-LT. This is similar to the other studies (25-35%), even though it was in a different baseline dosing of MMF (1.0 g/day vs. 2.0-3.0 g/day).²⁵⁻²⁸

Among all the side effects in this study that caused additional MMF dose adjustment, leukopenia was most common (87.1%). Even though WBC count at the time of the start of MMF exceeded $3,500/mm^3$ in all patients, leukopenia occurred in 36.5%. The reason why the recipients in our study were very sensitive to leukopenia, even in a reduced dose of MMF and of tolerable condition before administration, maybe due to pre-LT existing leukopenia, partial graft of LDLT, and relatively lower total body weight in this population.⁸ Pre-LT WBC count had mean $4,540/mm^3$ (range, $1,180-21,200/mm^3$), and leukopenia (< $3,500/mm^3$) was present in 44.6% at pre-LT. This considerable portion of preoperative leukopenia may be due to portal hypertension from underlying liver cir-

rhosis.

With respect to safety and efficacy, under tacrolimus based immunosuppression in adult LDLT, Yilmaz et al. reported the biopsy proven ACR as 11% for 107 recipients within the first year after LT,²⁹ and Sugawara et al. reported that as 45% for 100 recipients within the first three months after LT.³⁰ In our study, the overall biopsy-proven ACR was present in 2.7% of the cases, and this is excellent result compared to that of others. The reason for this excellent outcome may be due to the following characteristics in our cohort: 1) hepatitis B-virus related liver cirrhosis dominant baseline (75.7% in validating study)^{31,32}; 2) selected patients with pre-LT normal renal function (100.0%); 3) low Model for End-stage Liver Disease score (mean, 15.32 ± 8.23); 4) almost all were of elective LDLT (97.3%). In a similar cohort in our institute, we had reported that the 1-year survival rate was present in 96.0% and the graft survival in 99.0%, even with different immunosuppressant base.³³ Additionally, a relatively small sample size was one of the limitations of this study. For evaluation of the long-term outcome and renal preserving effects of immunosuppression of reduced dose of MMF combined with low trough level of tacrolimus in LT, long-term follow-up and more studies should be continued, henceforward.

In conclusion, a reduced dose of MMF was pharmacokinetically low compared to the existing recommended therapeutic range in early post-LDLT. Even with the low level of MPA AUC_{0-12} , there was no biopsy-proven ACR, graft loss, or death of patients under immunosuppression combined with low trough level of tacrolimus within the first year after LDLT. Even with the initially reduced dose of MMF, dose adjustment was frequently needed due to the side effects. After all, reduced dose of MMF combined with low levels of tacrolimus was feasible and safe with an excellent short-term outcome in hepatitis B virus dominant adult LDLT. More long-term follow-up and larger-scale prospective studies for this in LT are needed in the future.

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Conflicts of Interest

The authors have no conflicts to disclose.

REFERENCES

1. Pageaux GP, Rostaing L, Calmus Y, Duvoux C, Vanlemmens C, Hardgwissen J, et al. Mycophenolate mofetil in combination with reduction of calcineurin inhibitors for chronic renal dysfunction after liver transplantation. *Liver Transpl* 2006;12:1755-1760.
2. Arns W, Cibrik DM, Walker RG, Mourad G, Budde K, Mueller EA, et al. Therapeutic drug monitoring of mycophenolic acid in solid organ transplant patients treated with mycophenolate mofetil: review of the literature. *Transplantation* 2006;82:1004-1012.
3. Hwang S, Lee SG, Ahn CS, Kim KH, Moon DB, Ha TY, et al. A clinical assessment of mycophenolate drug monitoring after liver transplantation. *Clin Transplant* 2010;24:E35-42.
4. Kaplan B. Mycophenolic acid trough level monitoring in solid organ transplant recipients treated with mycophenolate mofetil: association with clinical outcome. *Curr Med Res Opin* 2006;22:2355-2364.
5. Premaud A, Le Meur Y, Debord J, Szelag JC, Rousseau A, Hoizey G, et al. Maximum a posteriori bayesian estimation of mycophenolic acid pharmacokinetics in renal transplant recipients at different postgrafting periods. *Ther Drug Monit* 2005;27:354-361.
6. Tredger JM, Brown NW, Adams J, Gonde CE, Dhawan A, Rela M, et al. Monitoring mycophenolate in liver transplant recipients: toward a therapeutic range. *Liver Transpl* 2004;10:492-502.
7. Tett SE, Saint-Marcoux F, Staatz CE, Brunet M, Vinks AA, Miura M, et al. Mycophenolate, clinical pharmacokinetics, formulations, and methods for assessing drug exposure. *Transplant Rev (Orlando)* 2011;25:47-57.
8. Yau WP, Vathsala A, Lou HX, Chan E. Is a standard fixed dose of mycophenolate mofetil ideal for all patients? *Nephrol Dial Transplant* 2007;22:3638-3645.
9. Suhail SM, Vathsala A, Lou HX, Woo KT. Safety and efficacy of mycophenolate mofetil for prophylaxis in Asian renal transplant recipients. *Transplant Proc* 2000;32:1757-1758.
10. Naito T. [Optimal immunosuppressive therapy based on pharmacokinetics and pharmacodynamics of antimetabolites in clinical practice]. *Yakugaku Zasshi* 2010;130:1695-1700.
11. Clavien PA, Barkun J, de Oliveira ML, Vauthey JN, Dindo D, Schulick RD, et al. The Clavien-Dindo classification of surgical complications: five-year experience. *Ann Surg* 2009;250:187-196.
12. Banff schema for grading liver allograft rejection: an international consensus document. *Hepatology*. 1997;25:658-663.
13. A blinded, randomized clinical trial of mycophenolate mofetil for the prevention of acute rejection in cadaveric renal transplantation. The Tricontinental Mycophenolate Mofetil Renal Transplantation Study Group. *Transplantation* 1996;61:1029-1037.
14. van Gelder T, Le Meur Y, Shaw LM, Oellerich M, DeNofrio D, Holt C, et al. Therapeutic drug monitoring of mycophenolate mofetil in transplantation. *Ther Drug Monit* 2006;28:145-154.
15. Shaw LM, Figurski M, Milone MC, Trofe J, Bloom RD. Therapeutic drug monitoring of mycophenolic acid. *Clin J Am Soc Nephrol* 2007;2:1062-1072.
16. Lampon N, Tutor-Crespo MJ, Romero R, Tutor JC. Diagnostic efficiency of truncated area under the curve from 0 to 2 h (AUC₀₋₂) of mycophenolic acid in kidney transplant recipients receiving mycophenolate mofetil and concomitant tacrolimus. *Clin Chem Lab Med* 2011;49:1167-1170.
17. van Gelder T. Mycophenolate blood level monitoring: recent progress. *Am J Transplant* 2009;9:1495-1499.
18. Bruchet NK, Ensom MH. Limited sampling strategies for mycophenolic acid in solid organ transplantation: a systematic review. *Expert Opin Drug Metab Toxicol* 2009;5:1079-1097.
19. Le Meur Y1, Büchler M, Thierry A, Caillard S, Villemain F, Lavaud S, et al. Individualized mycophenolate mofetil dosing based on drug exposure significantly improves patient outcomes after renal transplantation. *Am J Transplant* 2007;7:2496-2503.
20. Barraclough KA, Isbel NM, Franklin ME, Lee KJ, Taylor PJ, Campbell SB, et al. Evaluation of limited sampling strategies for mycophenolic acid after mycophenolate mofetil intake in adult kidney transplant recipients. *Ther Drug Monit* 2010;32:723-733.
21. Brunet M, Cirera I, Martorell J, Vidal E, Millan O, Jimenez O, et al. Sequential determination of pharmacokinetics and pharmacodynamics of mycophenolic acid in liver transplant patients treated with mycophenolate mofetil. *Transplantation* 2006;81:541-546.
22. Knoll GA, MacDonald I, Khan A, Van Walraven C. Mycophenolate mofetil dose reduction and the risk of acute rejection after renal transplantation. *J Am Soc Nephrol* 2003;14:2381-2386.
23. Mourad M1, Malaise J, Chaib Eddour D, De Meyer M, König J, Schepers R, et al. Correlation of mycophenolic acid pharmacokinetic parameters with side effects in kidney transplant patients treated with mycophenolate mofetil. *Clin Chem* 2001;47:88-94.
24. Pelletier RP, Akin B, Henry ML, Bumgardner GL, Elkhammas EA, Rajab A, et al. The impact of mycophenolate mofetil dosing patterns on clinical outcome after renal transplantation. *Clin Transplant* 2003;17:200-205.
25. Dumortier J, Guillaud O, Pittau G, Salandre J, Adham M, Scoazec JY, et al. Introduction of mycophenolate mofetil in maintenance liver transplant recipients: what can we expect? Results of a 10-year experience. *Transplant Proc* 2010;42:2602-2606.
26. Pfitzmann R, Klupp J, Langrehr JM, Uhl M, Neuhaus R, Settmacher U, et al. Mycophenolatemofetil for immunosuppression after liver transplantation: a follow-up study of 191 patients. *Transplantation* 2003;76:130-136.

27. Sollinger HW. Mycophenolates in transplantation. *Clin Transplant* 2004;18:485-492.
28. Hardinger KL, Brennan DC, Lowell J, Schnitzler MA. Long-term outcome of gastrointestinal complications in renal transplant patients treated with mycophenolate mofetil. *Transpl Int* 2004;17:609-616.
29. Banff Working Group, Demetris AJ, Adeyi O, Bellamy CO, Clouston A, Czaja A, et al. Liver biopsy interpretation for causes of late liver allograft dysfunction. *Hepatology* 2006;44:489-501.
30. Imamura H, Matsuyama Y, Tanaka E, Ohkubo T, Hasegawa K, Miyagawa S, et al. Risk factors contributing to early and late phase intra-hepatic recurrence of hepatocellular carcinoma after hepatectomy. *J Hepatol* 2003;38:200-207.
31. Crespo G1, Mariño Z, Navasa M, Forns X. Viral hepatitis in liver transplantation. *Gastroenterology* 2012;142:1373-1383
32. Burra P, Germani G, Adam R, Karam V, Marzano A, Lampertico P, et al. Liver transplantation for HBV-related cirrhosis in Europe: an ELTR study on evolution and outcomes. *J Hepatol* 2013;58:287-296.
33. Yi NJ, Suh KS, Suh SW, Chang YR, Hong G, Yoo T, et al. Excellent outcome in 238 consecutive living donor liver transplantations using the right liver graft in a large volume single center. *World J Surg* 2013;37:1419-1429.