# Title: Investigation on the benefits of mycophenolate mofetil and therapeutic drug monitoring in the treatment of Japanese patients with lupus nephritis

Takayuki Katsuno[1,3], Takenori Ozaki[2], Takaya Ozeki[1], Asaka Hachiya[1], Hangsoo Kim[1], Noritoshi Kato[1], Takuji Ishimoto[1], Sawako Kato[1], Tomoki Kosugi[1], Naotake Tsuboi[1], Masashi Mizuno[1], Yasuhiko Ito[3], and Shoichi Maruyama[1]

[1] Department of Nephrology, Nagoya University Graduate School of Medicine, Nagoya, Japan

[2] Department of Internal Medicine, Eijinkai Sato Hospital, Konan, Japan

[3] Department of Nephrology and Rheumatology, Aichi Medical University, Nagakute, Japan

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**Corresponding author:**

Takayuki Katsuno, MD, PhD

Department of Nephrology Nagoya University Graduate School of Medicine

65 Tsurumai-cho, Showa-ku Nagoya 466-8550, Japan

Phone: +81-52-744-2192 Fax: +81-52-744-2209

Email: katsu11@med.nagoya-u.ac.jp

# Abstract

***Background***

Mycophenolate mofetil (MMF) is recommended as a first-line immunosuppressant to treat lupus nephritis (LN). Prognosis and therapeutic response in LN are known to vary depending on race. We investigated the benefits of MMF and therapeutic drug monitoring (TDM) in the treatment of Japanese LN patients.

***Methods***

In this retrospective cohort study, a total of 20 patients with LN who started MMF treatment were included. Clinical data were collected regularly after MMF administration. We evaluated complete remission (CR) rate as the primary outcome. Predictors of CR were identified using univariate and multivariate analysis. In the research of TDM, the correlation with the area under the curve (AUC) was analyzed at MMF dose, single-point value, treatment response and adverse events.

***Results***

Overall, 70% of cases showed CR; both flare-ups and refractory cases had favorable results. Cases of LN with nephrotic syndrome (NS) or class III/IV+V showed a

significantly lower CR rate ($p < 0.005$). The ratio of maintaining CR after MMF therapy was as high as 85.7%. In multivariate analysis, NS was an independent negative predictor of CR (HR: 0.09, 95% confidence interval: 0.01–0.81; $p = 0.03$). The relationship between AUC and MMF dose was low, and AUC correlated with trough level ($r = 0.73$). AUC tended to be high in the treatment responder ($p = 0.09$), but did not correlate with adverse events of infection ($p = 0.92$).

***Conclusion***

MMF is a beneficial treatment option for Japanese LN patients, and further investigation on TDM-based therapy is needed.

# Introduction

Lupus nephritis (LN) is a major complication of systemic lupus erythematosus (SLE) that requires aggressive immunosuppressive therapy. In the treatment of LN, cyclophosphamide has played a central role. In fact, intravenous cyclophosphamide (IVCY) was the first immunosuppressive agent demonstrated by randomized controlled trial (RCT) to be superior to steroid-alone treatment [1]. However, administration to young patients and long-term use are not recommended for severe side effects. Mycophenolate mofetil (MMF) is effective as an immunosuppressant in organ transplantation, and its efficacy has also been demonstrated in the treatment of LN. Specifically, in a large-scale RCT (Aspreva Lupus Management Study; ALMS), the therapeutic response and safety of MMF were shown to be comparable to those of IVCY [10]. In addition, recent guidelines recommended MMF as a first-line drug in the treatment of LN [2, 3].

Race and ethnicity have been shown to have prognostic importance in LN [4]. In this regard, since few reports have focused on the efficacy of MMF in Japanese patients with LN, more clinical research is necessary.

Therapeutic drug monitoring (TDM) of MMF is common in kidney transplantation and can in fact reduce the risk of allograft rejection and treatment failure [5]. However, TDM has rarely been implemented in the treatment of LN. Therefore, further investigation into TDM in the treatment of LN using MMF is required. The aim of this study is to assess the therapeutic benefits and safety of MMF, as well as to investigate the utility of TDM in the treatment of Japanese patients with LN.

# Materials and methods

## Study design and population

This is a retrospective cohort study conducted in a single medical institution. The study subjects comprised 31 LN patients who started MMF treatment at Nagoya University Hospital between December 2006 and January 2016. SLE was diagnosed according to the American College of Rheumatology classification criteria [6]. The exclusion criteria for analysis were as follows: (1) observation period of less than 1 year, (2) urinary protein less than 0.5 g/24 hours or g/g creatinine (Cre) at the initiation of MMF, (3) concurrent use of biological agents, and (4) MMF treatment for disease other than SLE. According to these criteria, 11 patients were excluded, with the remaining 20 patients ultimately included. The study protocol was approved by the Standards of Official Conduct Committee of Nagoya University Hospital (approval number: 2017-0086).

## Clinical data collection and renal pathological finding

Baseline data, including clinical characteristics at the start of MMF treatment,

medical history, and renal pathological findings, were obtained from the hospital records. Pathological diagnosis of LN was based on the classification criteria of the International Society of Nephrology/Renal Pathology Society (ISN/RPS) [7]. In the case of clinical parameters, namely serum C3 and C4 levels, anti-DNA antibody levels, estimated glomerular filtration rate (eGFR), urinary protein creatinine ratio (uPCR), and the dosages of prednisolone (PSL) and MMF, data collection was also carried out 3, 6, and 12 months after MMF initiation, and the therapeutic effect was verified. In addition to the baseline, SLE disease activity index (SLEDAI) was scored again after 12 months.

## Renal outcome analysis and definition of terms

The primary outcome of LN treatment was complete remission (CR). Differences in CR rate in terms of clinical findings, renal pathology, and treatment methods were investigated, and predictors of CR were identified. Furthermore, maintenance of CR after MMF therapy was also evaluated. CR was defined as the return of serum creatinine to its previous baseline, plus a decline in the uPCR to less than 0.5 g/g Cre, in accordance with the Kidney Disease Improving Global Outcomes

(KDIGO) Clinical Practice Guideline for Glomerulonephritis [8]. When the baseline data were unknown in the initial cases, it was defined as CR that the serum creatinine was in the normal range. Renal flare was identified, as described in the ALMS trial [10].

## Treatment and therapeutic drug monitoring of MMF

MMF was orally administered twice daily every 12 hours. The initial dose was adjusted between 250 and 1500 mg according to renal function. Among them, 1000 mg was the most frequent, accounting for 70% of the total. Subsequently, the optimal dose of MMF was determined and maintained with the upper limit of 2000 mg in consideration of therapeutic response, side effect and blood concentration of MMF. Depending on the case, MMF was reduced in the maintenance phase after the treatment effect was observed.

TDM of MMF was carried out at various times, such as the remission induction and maintenance phase. Blood samples were collected at 0, 1, 3, and 6 hours after oral administration of MMF, and mycophenolic acid (MPA) concentrations were measured using Liquid Chromatography/Mass Spectrometry in a total of 72 samples

collected from 12 patients. The area under the plasma concentration-time curve (AUC) was calculated according to a previously reported method [9]. We investigated whether the AUC correlated with MMF dose and data at single-points. Furthermore, the relation between therapeutic response or infectious disease and AUC was verified, limited to 10 cases in which AUC was calculated at each maximum MMF dose.

## Statistical analysis

Clinical data were shown as either medians with interquartile ranges (IQRs) or numbers with percentages (%). Differences between groups were analyzed using the Wilcoxon rank-sum test (for continuous variables) or Fisher's exact test (for categorical variables). To analyze changes in clinical parameters, the paired t-test was adopted. We used the Kaplan–Meier method to evaluate the cumulative CR rate after MMF therapy, and the log-rank test to compare differences between the two groups. To determine factors predicting CR, we used the univariate and multivariate Cox proportional hazards model. The results were expressed as hazard ratios (HRs) with 95% confidence intervals (CIs). The proportional hazards assumption for covariates was tested using scaled

Schoenfeld residuals. With regards to the TDM data, the correlation was analyzed using the Pearson product-moment correlation coefficient. Statistical significance was set at a p-value of $< 0.05$. All statistical tests were performed using Stata version 14.0 (Stata Corp LLC, College Station, TX, USA)

# Results

## Patient characteristics

The patients' baseline clinicopathological findings at the start of MMF treatment are summarized in Tables 1 and 2. The patients were divided into two groups: initial treatment cases and flare cases. In the flare group, patients tended to be older. Disease activity, indicated by complement titer, anti-DNA antibody, and SLEDAI, was confirmed to be higher in the initial treatment group. Regarding treatment, a smaller amount of PSL was administered in the flare group ($p = 0.047$), and significantly more patients had received immunosuppressive drugs previously ($p = 0.004$). With regards to pathological features, LN categorized as class IV was only observed in the initial treatment group; cases of membranous LN (class V) were more frequent in the flare group.

## Clinical parameters

The serum C3 and C4 levels, anti-DNA antibody levels, and SLEDAI had improved significantly after 12 months of MMF treatment. Specifically, the median

serum C3 and C4 had increased from 65.6 mg/dL (IQR: 53.9–69.3 mg/dL) and 12.1 mg/dL (IQR: 6.5–17.3 mg/dL), respectively, at baseline to 96.6 mg/dL (IQR: 83.8–106.1 mg/dL) and 19.0 mg/dL (IQR: 17.3–22.4 mg/dL), respectively, after 12 months ($p < 0.01$). The median anti-DNA antibody levels decreased from 17.3 IU/mL (IQR: 8.1–266.0 mg/dL IU/mL) at baseline to 5.4 IU/mL (IQR: 2.8–11.1 IU/mL) at 12 months ($p < 0.05$). The SLEDAI, whose median value was 12 (IQR: 8–16) at baseline, decreased to 4 (IQR: 2–6) after 12 months ($p < 0.01$). The median urinary protein at baseline was 2.62 g/g Cre (IQR: 1.34–5.51 g/g Cre); after 12 months it decreased to 0.19 g/g Cre (IQR: 0.06–1.00 g/g Cre; $p < 0.01$). PSL dosage decreased from a median of 40 mg/day (IQR: 30–50 mg/day) at baseline to 9.5 mg/day (IQR: 7–10 mg/day) after 12 months ($p < 0.01$). The median MMF dosage at baseline was 1000 mg/day (IQR: 1000–1000 mg/day), and it did not change significantly after 12 months (Fig. 1).

## Renal outcome

Initially, survival analysis was performed with CR as the primary outcome in the 20 cases of active LN. The Kaplan–Meier curves are shown in Fig. 2. During the

observation period (median: of 21.8 months, IQR: 13.4–53.6 months), 14 (70%) patients showed CR. The median time to CR was 51 days (IQR: 20–161 days). Among the 13 patients who had already been treated using other immunosuppressive drugs prior to starting MMF treatment, eight (61.5%) achieved CR. The median time to CR was 105 days (IQR: 28–252 days). Subsequently, the subjects were divided into two groups, and the cumulative CR rates were compared (Fig. 3). CR was found in nine out of 11 (81.8%) patients in the initial treatment group, and in five out of nine (55.6%) patients in the flare group. There was no significant difference between the groups in this regard ($p = 0.27$). In a comparison between patients with and without NS, 12 of 13 (92.3%) non-NS patients showed CR, while only two of seven (28.6%) patients with NS achieved CR. The CR rate was significantly lower in the NS group ($p < 0.005$). Concerning the correlation between CR rate and membranous (class V) LN, all six patients with class III/IV LN showed CR, whereas six of 10 (60%) with class V LN had CR. Patients with class V presented a significantly lower CR rate ($p < 0.005$). In the initial treatment group, CR was observed in six out of seven (85.7%) patients who were treated using a combination therapy of PSL and MMF. There was no significant

difference in CR rate among patients treated using a multi-target therapy that added a calcineurin inhibitor (Fig. 4).

We further investigated the maintenance of CR achieved by MMF therapy in 14 patients (Fig. 5). Twelve patients (85.7%) maintained CR during the follow-up period, which had a median duration of 34.3 months (IQR: 10.2–36 months). Two patients who could not maintain CR were in the initial treatment group, and their individual times to failure were 5.8 months and 31.6 months. All patients in the flare group maintained CR.

## Predictors of CR

Univariate analysis indicated that high age (HR: 0.64, 95% CI: 0.45–0.92; p = 0.01), NS (HR: 0.14, 95% CI: 0.03–0.64; p = 0.01), and membranous (class V) LN (HR: 0.19, 95% CI: 0.05–0.69; p = 0.01) were negative predictors of CR. Furthermore, multivariate analysis revealed that NS was an independent negative predictor (HR: 0.09, 95% CI: 0.01–0.81; p = 0.03; Table 3).

## Analysis of TDM

The MMF dose at the time of TDM was 1375 mg/day (IQR: 1000–1500 mg/day) as a median value. The median PSL dosage was 25 mg/day (IQR: 7–35 mg/day). We examined the correlation between 12-hour AUC—calculated from the plasma concentration of MPA (MPA-AUC 0–12)—and MMF dosage or single-point value (Fig. 6). The correlation between MPA-AUC 0–12 and MMF dosage was not strong (r = 0.53). The plasma concentrations of MPA 0, 1, 3, and 6 hours after oral administration correlated with the MPA-AUC 0–12. Among them, trough values were significantly correlated with MPA-AUC 0–12 (r = 0.73). Fig. 7 shows an analysis of the relationship between MPA-AUC 0–12 and therapeutic effect or adverse events of infection including cytomegalovirus (CMV) reactivation, herpes zoster and pneumonia in 10 patients. In the responder group including six patients with CR, the median value of AUC 0–12 was 52.6 mg·hours/L (IQR: 51.2–53.2 mg·hours/L). Conversely, the median AUC 0–12 of the non-responder group consisting of four patients was 43.5 mg·hours/L (IQR: 41.0–45.6 mg h/L), which was lower than that of the responder group (p = 0.09). Infectious events were observed in five out of 10

patients. The median AUC 0–12 was 51.2 mg·hours/L (IQR: 45.2–52.5 mg h/L) in patients with infection and 46.1 mg·hours/L (IQR: 40.2–53.2 mg h/L) without infection ($p = 0.92$). There was no correlation between AUC 0–12 and infectious events.

## Adverse events

The adverse events are summarized in Table 4. The major events were infections: three cases of herpes zoster and four cases of infection requiring hospitalization were observed. Regarding CMV infection, seven out of 17 (41.2%) patients showed CMV reactivation. The median period to reactivation was 26 days (IQR: 20–63), and most cases developed within 3 months of MMF initiation. There were no cases of CMV infection with severe organ damage. Leukocytopenia and gastrointestinal symptoms occurred with low frequency, with each complaint comprising only one case.

# Discussion

Several RCTs have compared MMF with oral or intravenous CY as an induction therapy for LN. Specifically, MMF demonstrated comparable or superior clinical efficacy [10, 11]. Based on the results of these trials, MMF is a beneficial treatment option, taking remission rate, flare rate, and adverse effects into consideration.

In this cohort study, CR was observed in 70% of all cases, which was a better outcome than in previous RCTs [10, 11]. There may be a number of reasons for these different CR rates. In the present study, the median observation period was relatively long. The baseline renal function has not decreased, and the composition of the renal pathology was different. Differences in the definitions of CR may have influenced. The prognosis of LN, as well as the therapeutic response of the condition to immunosuppressive drugs, are known to be influenced by race, and it seems that treatment in Japanese patients is related to favorable outcomes. The CR was achieved in eight out of 13 (61.5%) patients with LN who had already been treated using immunosuppressants other than MMF. In the comparison between initial and flare cases, there was no significant difference in cumulative CR rate. Although several RCTs have

compared induction therapies for LN, as mentioned above, few reports have discussed the effects of alterations to treatment modalities. The present study indicated that there are cases in which CR has been achieved by treatment with MMF in flare and refractory cases.

According to one survey, NS is regarded as an unfavorable prognostic factor in LN [12]. In this cohort, the CR rate in patients exhibiting NS at baseline was significantly lower, and NS was an independent negative predictor of CR in the multivariate analysis. In the therapeutic strategy of LN with NS, there is a possibility of causing excessive immunosuppression when targeting CR. In some cases, it may be reasonable to set treatment intensity aiming for partial remission.

The impact of histological patterns on the treatment response of LN patients has also been reported [13]. Patients at the overlap of class V with classes III or IV showed poor therapeutic response. In a prospective study, Bao et al. carried out multi-target therapy by combining PSL, tacrolimus, and MMF in class IV+V LN, demonstrating that this treatment approach was superior to IVCY alone [14]. Even in the present study, overlapping class V LN presented poor results. However, this

267 statistical difference seems to have been largely influenced by the time required for
268 remission. Indeed, the CR rate in patients with class III/IV+V LN was 60% (six out of
269 10 patients) in the present study, and although the observation period was different, our
270 results were comparable to those of multi-target therapy from China. Considering the
271 results of multivariate analysis, our research suggested that the histopathological finding
272 with class V may affect the time to CR, but not correlate the CR rate.

273 LN flare is reported to correlate with a risk of progressive chronic kidney
274 disease [15]. In maintenance therapy, two landmark RCTs have been conducted: the
275 ALMS maintenance trial [16] and the MAINTAIN nephritis trial [17]. These trials
276 reported on the efficacy of MMF in maintenance therapy. In this research, maintenance
277 therapy using MMF showed favorable results. This result was similar to that of the
278 ALMS maintenance trial, indicating that MMF is superior to azathioprine.

279 The data regarding TDM in patients with LN are limited. Shaw et al. reported
280 that there was high between-patient variability of MPA-AUC in organ transplant
281 patients [18]. This variability was also identified in a study involving 71 SLE patients
282 [19]. Therefore, TDM seems to be important in the treatment of LN. There have been

two reports investigating TDM in LN patients [20, 21]; both showed similar results. MPA-AUC correlated with therapeutic response and AUC 0–12 level above 45 mg·hours/L can precisely predict favorable results. Furthermore, the same reports demonstrated that trough value and MPA-AUC were significantly correlated. These findings contradict those in renal transplant recipients, which indicated weak correlations between trough value and MPA-AUC [22]. Regarding the correlation between trough value and MPA-AUC, the analysis results of TDM in our cohort study were similar to previous reports. On the other hand, MPA-AUC tended to be higher in the responder group, but statistically significant difference was not observed. Interestingly, at the dose of MMF in this study, no significant correlation was found between MPA-AUC and adverse events of infection. We confirmed individual disparities in drug absorption kinetics. In order to clarify the usefulness of TDM and application method of TDM data in the treatment of Japanese LN patients, further research is required for many cases.

The most common adverse events were infectious diseases. Among them, CMV antigenemia was frequently observed—most often within 3 months of the start of

MMF treatment. However, this may have been correlated with the high dosage PSL that was the concomitant medication. There were overwhelmingly few cases of hematopenia and gastrointestinal disorders. These results may be related to the dosage of MMF less than that of the major RCTs.

The foremost limitation of the present study was its small sample size. Other limitations are as follows: (1) it was not a comparative trial involving other immunosuppressive treatments such as IVCY; (2) the observation period differed from that of the major RCTs, and it was therefore hard to evaluate treatment efficacy uniformly; (3) patient background varied and relatively mild LN was also included in the study; (4) TDM of MMF was not performed in all cases; (5) considering enterohepatic circulation, the accuracy of the prediction formula of AUC 0–12 improves by measuring blood concentration 8 and 9 hours after administration of MMF, but it is not implemented in this study. Despite these limitations, our research showed that MMF is a beneficial treatment option for Japanese LN patients. Further investigations focusing on the optimum dose of MMF based on TDM, the treatment duration of MMF and concomitant medication are necessary.

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**Table 1**

**Baseline characteristics of lupus nephritis patients with MMF therapy**

Values are shown as either medians with interquartile ranges (IQRs) or numbers with percentages (%).

Abbreviations: BMI, body mass index; eGFR, estimated glomerular filtration rate; IgG, immunoglobulin G; MMF, mycophenolate mofetil; SLE, systemic lupus erythematosus; SLEDAI, systemic lupus erythematosus disease activity index; uPCR, urinary protein creatinine ratio

*$p < 0.05$ (initial treatment cases vs. flare cases)

**Table 2**

**Baseline data of treatment and renal pathology in the patients treated with MMF**

Values are shown as either medians with interquartile ranges (IQRs) or numbers with percentages (%).

Abbreviations: ARB, angiotensin receptor blocker; ACE, angiotensin-converting enzyme inhibitor; CyA, cyclosporine; ISN/RPS, International Society of

Nephrology/Renal Pathology Society; IVCY, intravenous cyclophosphamide; MMF, mycophenolate mofetil; MZR, mizoribine; PSL, prednisolone; TAC, tacrolimus

*$p < 0.05$ (initial treatment cases vs. flare cases)

**Table 3**

**Univariate and multivariate analysis for predictive factors of complete remission**

Abbreviations: BMI, body mass index; CI, confidence interval; eGFR, estimated glomerular filtration rate; HR, hazard ratio; SLEDAI, systemic lupus erythematosus disease activity index; uPCR, urinary protein creatinine ratio

*$p < 0.05$

**Table 4**

**Adverse events during observation period**

Abbreviations: CMV, cytomegalovirus; WBC, white blood cell

*The percentage was calculated in 17 patients whose CMV antigen levels were measured.

**Fig. 1 The dosages of PSL and MMF.**

The dosage of PSL decreased significantly with time. Conversely, the dosage of MMF remained largely unchanged.

Abbreviations: MMF, mycophenolate mofetil; PSL, prednisolone

*$p < 0.01$ (vs. baseline)

**Fig. 2 The CR rate of lupus nephritis.**

Kaplan–Meier curves show the cumulative CR rates in all patients, as well as in patients who had received other immunosuppressive drugs previously.

Abbreviations: CR, complete remission; MMF, mycophenolate mofetil

**Fig. 3 Comparative analysis of CR rate between the two groups.**

The CR rate was significantly lower in nephrotic cases and in class III/IV+V lupus nephritis ($p < 0.005$).

Abbreviations: CR, complete remission; NS, nephrotic syndrome

**Fig. 4 Comparison of CR rate in terms of treatment regimen.**

Combination therapy comprising PSL and MMF was not inferior to multi-target therapy in the initial treatment cases ($p = 0.27$).

Abbreviations: CR, complete remission; CyA, cyclosporine; PSL, prednisolone; MMF, mycophenolate mofetil; TAC, tacrolimus

**Fig. 5 Maintenance of CR after MMF treatment.**

Kaplan–Meier curves show the maintenance rates of CR in the initial treatment and flare group.

Abbreviation: CR, complete remission; MMF, mycophenolate mofetil

**Fig. 6 The correlation between MPA-AUC and MMF dosage or MPA concentration at single-point.** (A) Even with the same MMF dose, there was between-patient variability in MPA-AUC. (B) With regards to the relationships between each single-point measurement value (C0, C1, C3, and C6) and MPA-AUC, the correlation with the trough level (C0) was the strongest ($r = 0.73$).

Abbreviations: AUC, area under the plasma concentration-time curve; MMF,

mycophenolate mofetil; MPA, mycophenolic acid

**Fig. 7 The relationship between MPA-AUC and therapeutic response or infectious adverse events.**

MPA-AUC was higher in the treatment responder ($p = 0.09$), and not related to the onset of infections ($p = 0.92$).

Abbreviations: AUC, area under the plasma concentration-time curve; MPA, mycophenolic acid

# Compliance with Ethical Standards

## Conflict of interest

The Department of Nephrology, Nagoya University Graduate School of Medicine received research promotion grants from Otsuka Pharmaceutical Co, Kissei Pharmaceutical Co, Novartis Pharma K.K, Kowa Pharmaceutical Co, Chugai Pharmaceutical Co, Nippon Boehringer Ingelheim Co ., Ltd, Pfizer Japan Inc, Kyowa Hakko Kirin Co ., Ltd, Torii Pharmaceutical Co.,Ltd, Astellas Pharma Inc, MSD K.K, Daiichi Sankyo Company Limited, Takeda Pharmaceutical Company Limited, Bristol-Myers Squibb, Mitsubishi Tanabe Pharma Corporation, Sumitomo Dainippon Pharma Co., Ltd, Teijin Pharma Limited, and Mochida Pharmaceutical Co ., Ltd.

## Human and Animal Rights

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional research committee at which the studies were conducted (IRB approval number 2017-0086) and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

## Informed Consent

The ethical committee approved this retrospective cohort study without written informed consent, but informed consent was obtained from most patients at the time of renal biopsy.

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Table 1

## Baseline characteristics of lupus nephritis patients with MMF therapy

| | All cases (n=20) | Initial treatment cases (n=11) | Flare cases (n=9) | p-value |
|---|---|---|---|---|
| Age at SLE diagnosis (years) | 26 [20.5-48.5] | 23 [16-53] | 29 [25-48] | 0.57 |
| Age at the start of MMF (years) | 39.5 [22-53.5] | 27 [20-53] | 45 [41-54] | 0.15 |
| Gender, female, n (%) | 18 (90.0) | 9 (81.8) | 9 (100.0) | 0.29 |
| BMI ($kg/m^2$) | 19.6 [17.6-23.8] | 19.4 [17.8-21.9] | 21.2 [17.2-25.1] | 0.43 |
| Systolic blood pressure (mmHg) | 125.5 [117-134] | 125 [117-134] | 130 [118-134] | 0.62 |
| Diastolic blood pressure (mmHg) | 76 [69.5-82.5] | 74 [69-83] | 80 [70-82] | 0.73 |
| Hypertension, n (%) | 5 (25.0) | 3 (27.3) | 2 (22.2) | 0.60 |
| Diabetes mellitus, n (%) | 5 (25.0) | 4 (36.4) | 1 (11.1) | 0.22 |
| **Laboratory parameters** | | | | |
| White blood cell count (/μL) | 8200 [6850-11500] | 8100 [6700-13800] | 8400 [6900-9200] | 0.65 |
| Hemoglobin (g/dL) | 10.9 [10.2-12.0] | 10.2 [9.9-11.0] | 11.5 [10.8-12.4] | 0.03* |
| Platelet count ($x10^4/\mu L$) | 21.5 [17.8-27.4] | 20.6 [17.0-28.0] | 22.4 [18.2-26.7] | 0.68 |
| Serum albumin (g/dL) | 2.8 [2.3-3.4] | 2.7 [2.1-3.0] | 3.2 [2.8-3.4] | 0.16 |
| Total cholesterol (mg/dL) | 258 [243.5-276.5] | 252 [239-277] | 264 [244-276] | 0.91 |
| Serum creatinine (mg/dL) | 0.56 [0.46-0.83] | 0.54 [0.44-0.65] | 0.73 [0.50-0.84] | 0.36 |
| eGFR ($ml/min/1.73m^2$) | 92.0 [77.5-129.5] | 109.2 [91.9-131.5] | 77.5 [58.6-111.7] | 0.05 |
| Serum complement activity (CH50) (U/mL) | 36.3 [17.3-47.7] | 39.3 [12.5-49.2] | 36.3 [32.3-45.5] | 0.62 |
| Serum C3 (mg/dL) | 65.6 [53.9-69.3] | 60.4 [37.9-66.9] | 67.8 [65.3-75.1] | 0.03* |
| Serum C4 (mg/dL) | 12.1 [6.5-17.3] | 7.4 [5.7-16.4] | 13.6 [11.0-18.1] | 0.14 |
| Serum IgG (mg/dL) | 870 [488-1334] | 950 [451-2011] | 790 [630-1045] | 0.65 |
| Anti-DNA antibody (IU/mL) | 17.3 [8.1-266.0] | 87.4 [11.2-320.1] | 9.6 [8.1-17.3] | 0.11 |
| uPCR (g/gCre) | 2.62 [1.34-5.51] | 2.60 [1.58-7.91] | 4.58 [1.33-5.30] | 0.79 |
| Nephrotic syndrome, n (%) | 7 (35.0) | 3 (27.3) | 4 (44.4) | 0.37 |
| SLEDAI | 12 [8-16] | 16 [8-26] | 10 [8-12] | 0.11 |

Table 2

## Baseline data of treatment and renal pathology in the patients treated with MMF

| | All cases (n=20) | Initial treatment cases (n=11) | Flare cases (n=9) | p-value |
|---|---|---|---|---|
| **Treatment** | | | | |
| ARB, n (%) | 8 (40.0) | 3 (27.3) | 5 (55.6) | 0.21 |
| ACE, n (%) | 2 (10.0) | 1 (9.1) | 1 (11.1) | 0.71 |
| PSL dosage (mg/day) | 40 [30-50] | 45 [40-50] | 30 [25-50] | 0.047* |
| MMF dosage (mg/day) | 1000 [1000-1000] | 1000 [1000-1000] | 1000 [500-1000] | 0.11 |
| Prior treatment with immunosuppressive drugs, n (%) | 13 (65.0) | 4 (36.4) | 9 (100.0) | 0.004* |
| TAC, n (%) | 4 (20.0) | 1 (9.1) | 3 (33.3) | 0.22 |
| CyA, n (%) | 6 (30.0) | 2 (18.2) | 4 (44.4) | 0.22 |
| MZR, n (%) | 5 (25.0) | 1 (9.1) | 4 (44.4) | 0.10 |
| IVCY, n (%) | 1 (5.0) | 1 (9.1) | 0 (0.0) | 0.55 |
| **Renal pathology (ISN/RPS classification)** | | | | |
| Class II, n (%) | 1 (5.0) | 0 (0.0) | 1 (11.1) | 0.45 |
| Class III, n (%) | 2 (10.0) | 1 (9.1) | 1 (11.1) | 0.71 |
| Class IV, n (%) | 4 (20.0) | 4 (36.4) | 0 (0.0) | 0.07 |
| Class III+V, n (%) | 5 (25.0) | 1 (9.1) | 4 (44.4) | 0.10 |
| Class IV+V, n (%) | 5 (25.0) | 2 (18.2) | 3 (33.3) | 0.40 |
| Unknown, n (%) | 1 (5.0) | 1 (9.1) | 0 (0.0) | |
| Not performed, n (%) | 2 (10.0) | 2 (18.2) | 0 (0.0) | |

Table 3

**Univariate and multivariate analysis for predictive factors of complete remission**

| Parameters | Univariate | | Multivariate | |
|---|---|---|---|---|
| | HR [95%CI] | p-value | HR [95%CI] | p-value |
| Age (every 10 years) | 0.64 [0.45-0.92] | 0.01* | – | – |
| Gender (male) | 1.23 [0.27-5.68] | 0.79 | – | – |
| BMI (kg/m$^2$) | 0.88 [0.75-1.04] | 0.13 | – | – |
| Initial treatment cases | 1.88 [0.62-5.68] | 0.27 | – | – |
| Flare cases | 0.53 [0.18-1.61] | 0.27 | – | – |
| Prior treatment with immunosuppressive drugs | 0.45 [0.15-1.32] | 0.14 | – | – |
| Serum creatinine (mg/dL) | 0.62 [0.21-1.85] | 0.39 | – | – |
| eGFR (ml/min/1.73m$^2$) | 1.01 [1.00-1.03] | 0.08 | – | – |
| Serum C3 (mg/dL) | 0.99 [0.96-1.02] | 0.41 | – | – |
| Serum C4 (mg/dL) | 0.93 [0.85-1.01] | 0.08 | – | – |
| Anti-DNA antibody (IU/mL) | 1.00 [1.00-1.01] | 0.01* | – | – |
| uPCR (g/gCre) | 0.88 [0.72-1.07] | 0.21 | – | – |
| Nephrotic syndrome | 0.14 [0.03-0.64] | 0.01* | 0.09 [0.01-0.81] | 0.03* |
| SLEDAI | 1.07 [1.01-1.15] | 0.03* | – | – |
| Renal pathology (Class V) | 0.19 [0.05-0.69] | 0.01* | 0.48 [0.12-1.86] | 0.29 |

Table 4

**Adverse events during observation period**

| | n (%) |
|---|---|
| Death | 0 (0.0) |
| Infection | |
| CMV antigenemia | 7 (41.2)* |
| Herpes zoster | 3 (15.0) |
| Respiratory infection requiring hospitalization | 3 (15.0) |
| Infectious enteritis requiring hospitalization | 1 (5.0) |
| Severe leukopenia, WBC count less than 3000/μL | 1 (5.0) |
| Gastrointestinal symptom (diarrhea and/or nausea) | 1 (5.0) |

Fig. 1

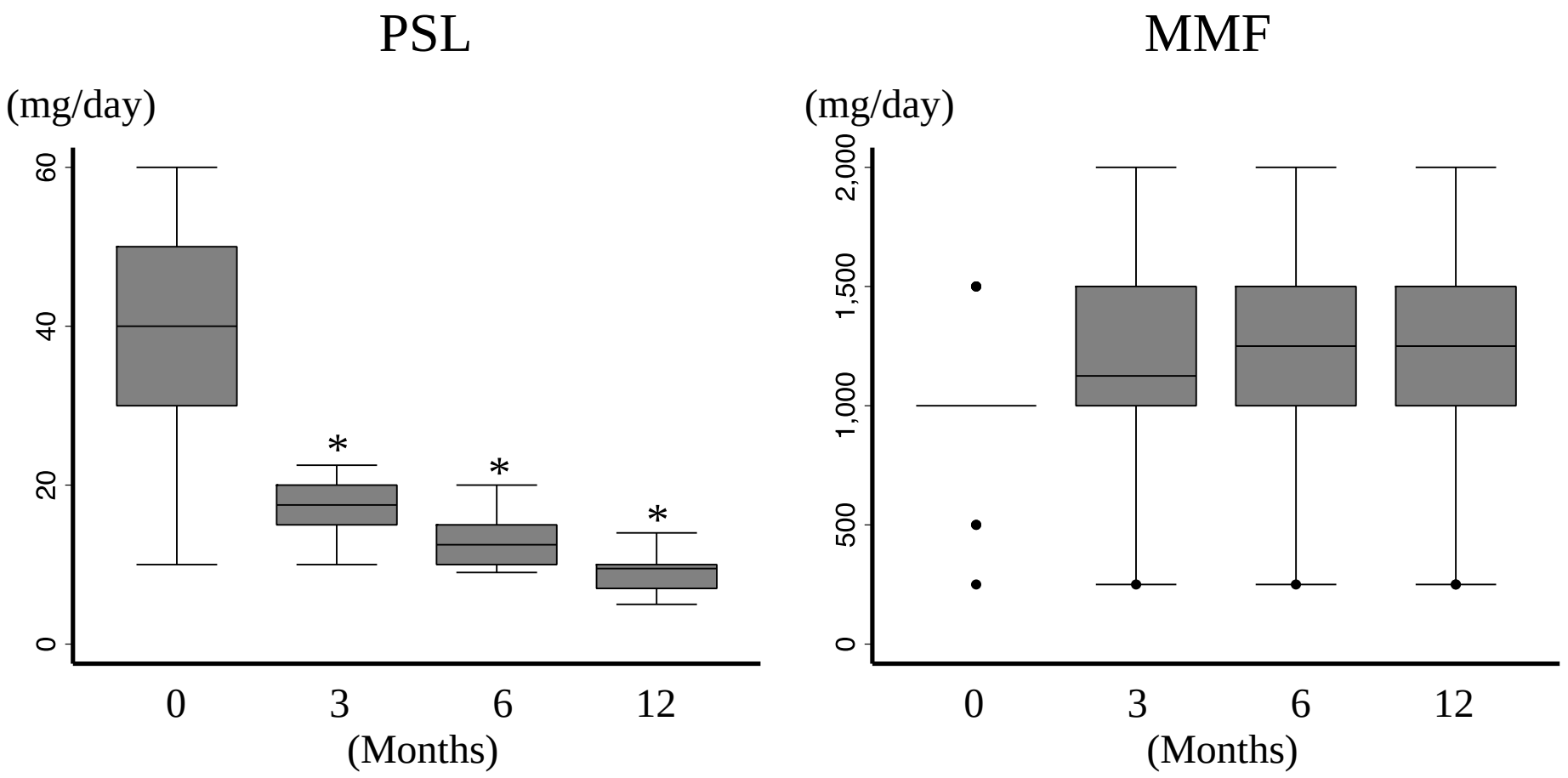
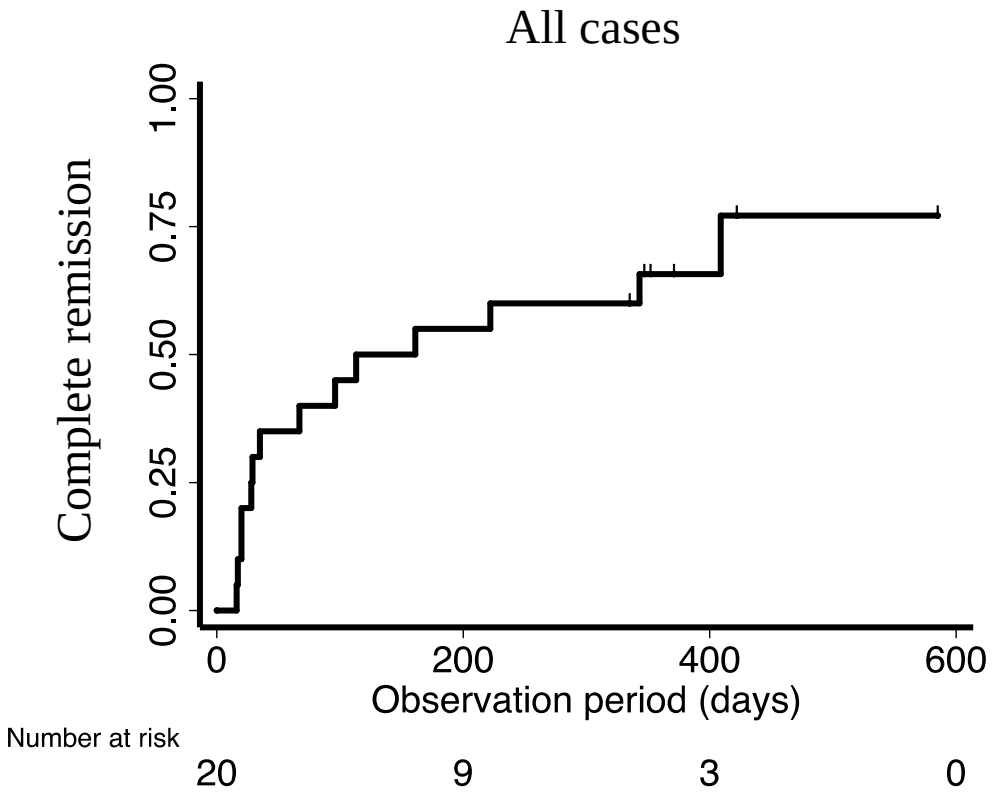

PSL
(mg/day)
60
40
20
0
*
*
*
0
3
6
12
(Months)
MMF
(mg/day)
2,000
1,500
1,000
500
0
0
3
6
12
(Months)

Fig. 2


All cases
Complete remission
0.00
0.25
0.50
0.75
1.00
0
200
400
600
Observation period (days)
Number at risk
20
9
3
0

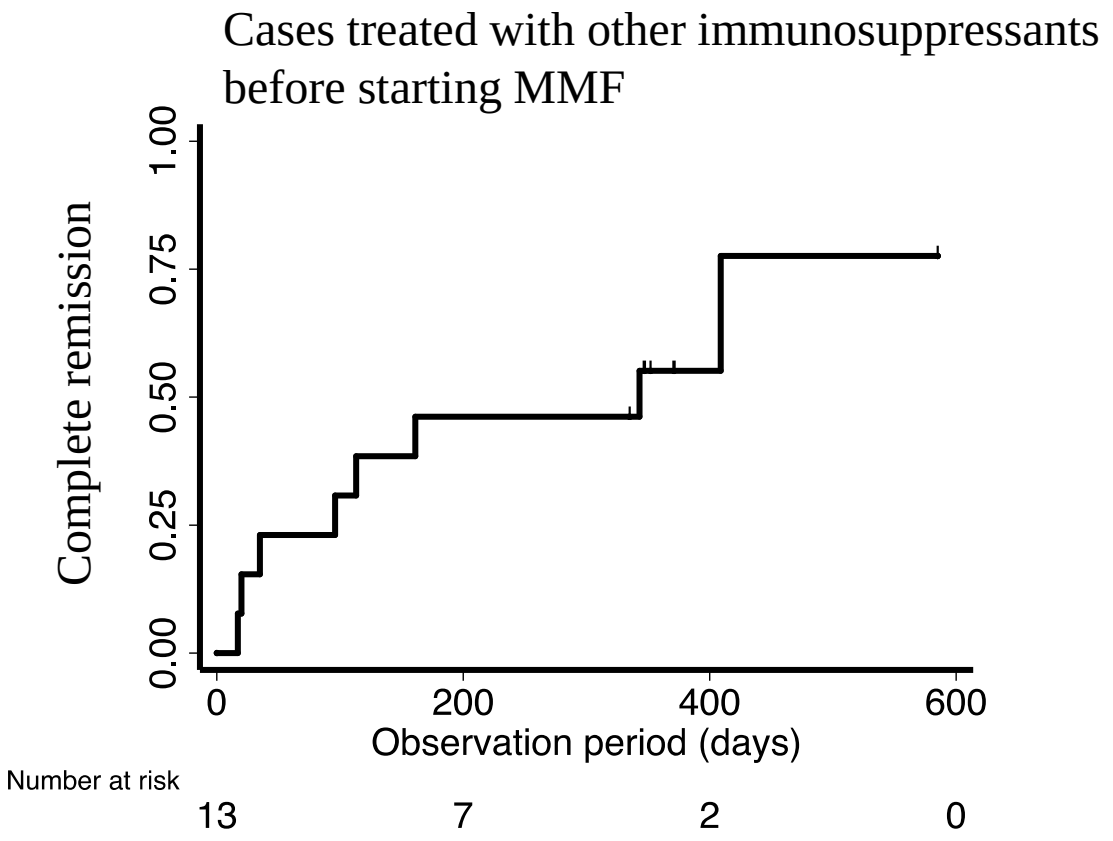
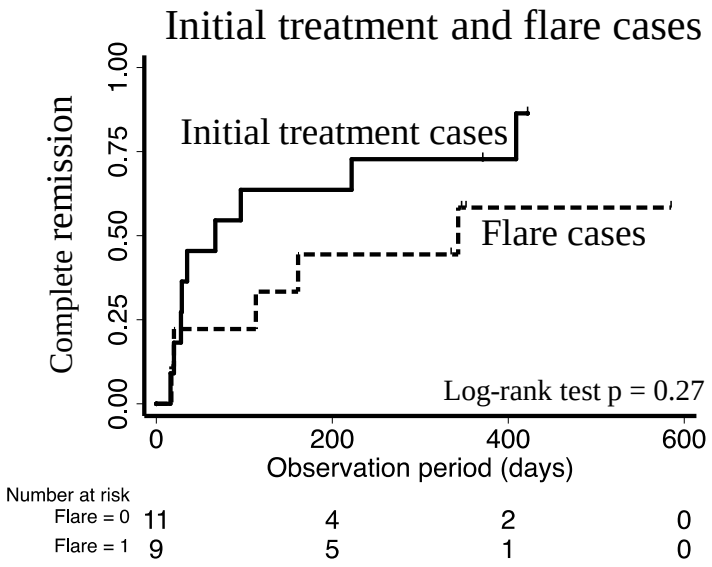
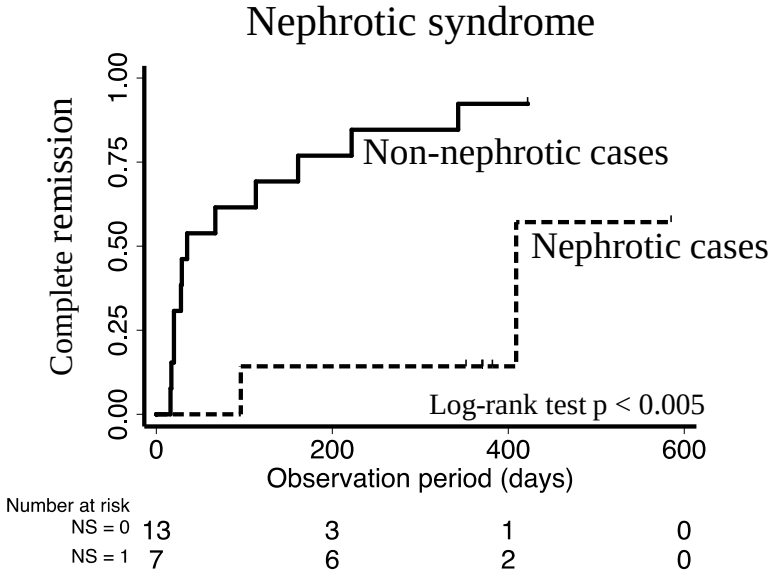

Cases treated with other immunosuppressants before starting MMF
Complete remission
0.00
0.25
0.50
0.75
1.00
0
200
400
600
Observation period (days)
Number at risk
13
7
2
0

Fig. 3


Initial treatment and flare cases
Complete remission
0.00
0.25
0.50
0.75
1.00
Initial treatment cases
Flare cases
Log-rank test p = 0.27
0
200
400
600
Observation period (days)
Number at risk
Flare = 0 11 4 2 0
Flare = 1 9 5 1 0


Nephrotic syndrome
Complete remission
0.00
0.25
0.50
0.75
1.00
Non-nephrotic cases
Nephrotic cases
Log-rank test p < 0.005
0
200
400
600
Observation period (days)
Number at risk
NS = 0 13 3 1 0
NS = 1 7 6 2 0

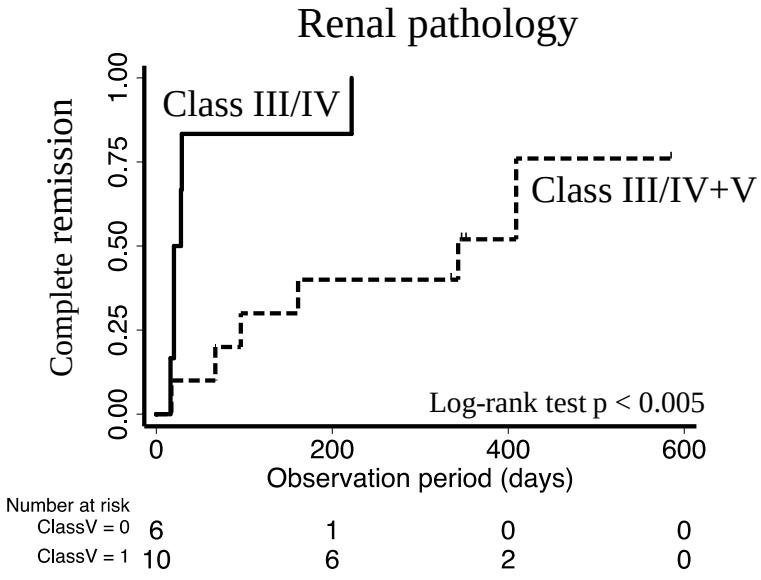

Renal pathology
Complete remission
0.00
0.25
0.50
0.75
1.00
Class III/IV
Class III/IV+V
Log-rank test p < 0.005
0
200
400
600
Observation period (days)
Number at risk
ClassV = 0 6 1 0 0
ClassV = 1 10 6 2 0

Fig. 4

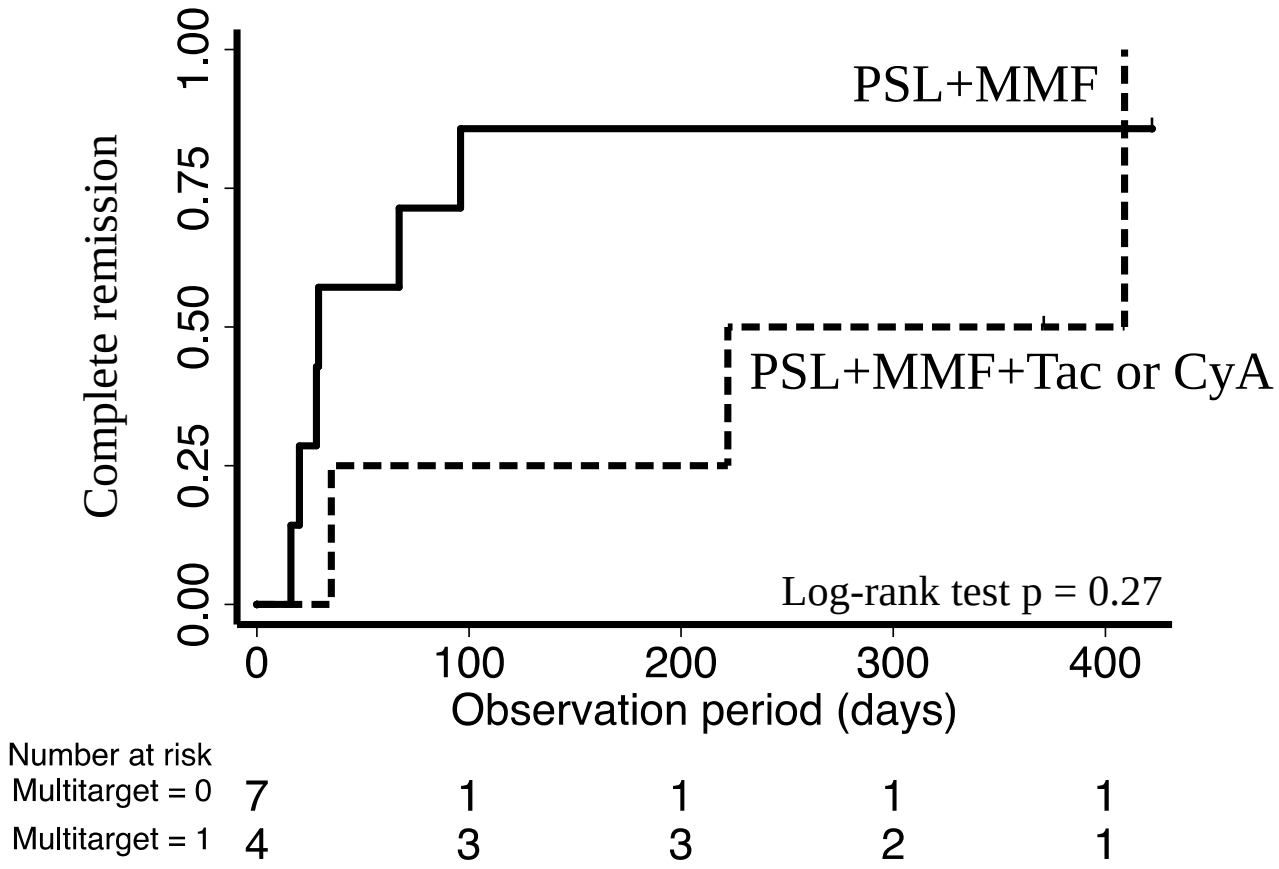

Complete remission
0.00
0.25
0.50
0.75
1.00
PSL+MMF
PSL+MMF+Tac or CyA
Log-rank test p = 0.27
0
100
200
300
400
Observation period (days)
Number at risk
Multitarget = 0
7
1
1
1
1
Multitarget = 1
4
3
3
2
1

Fig. 5

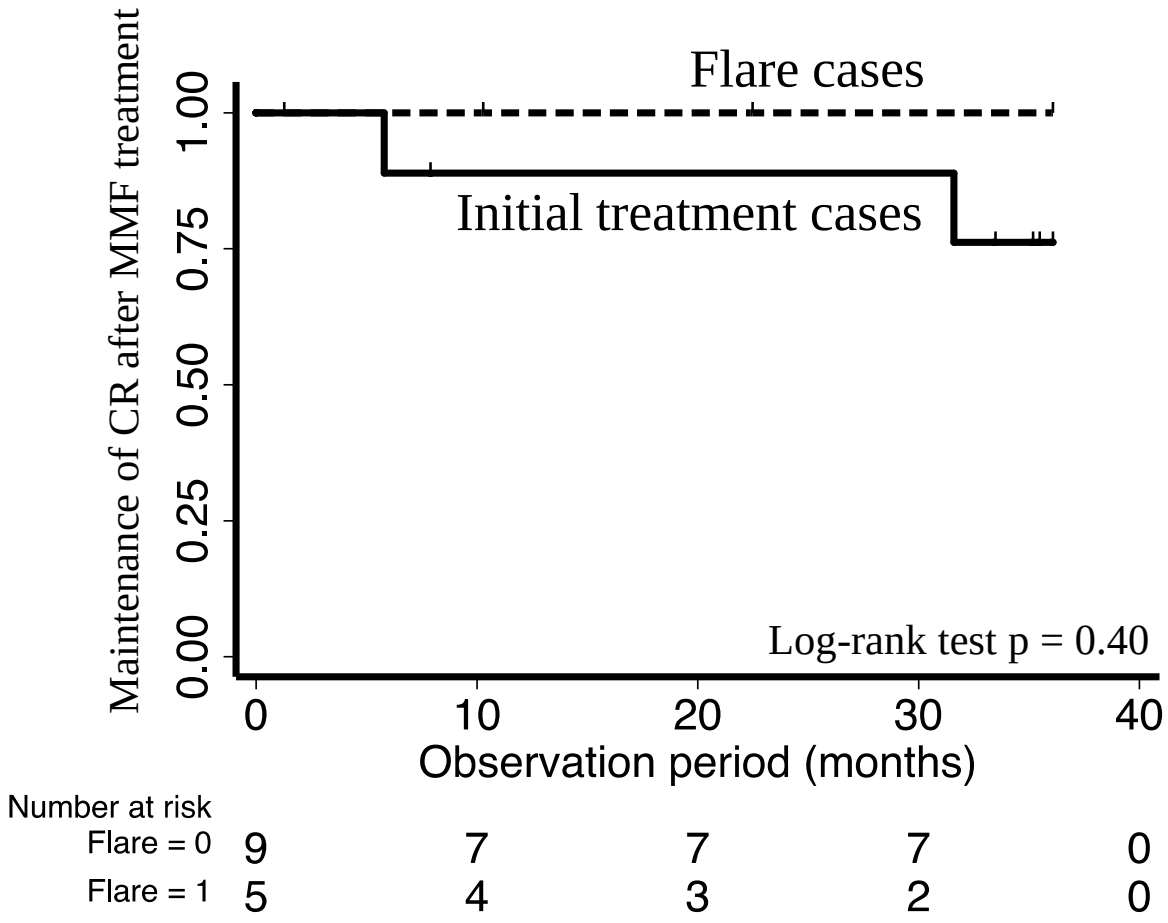
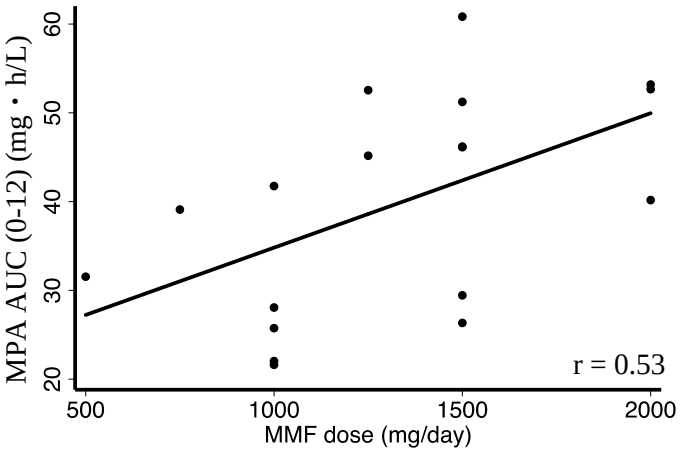
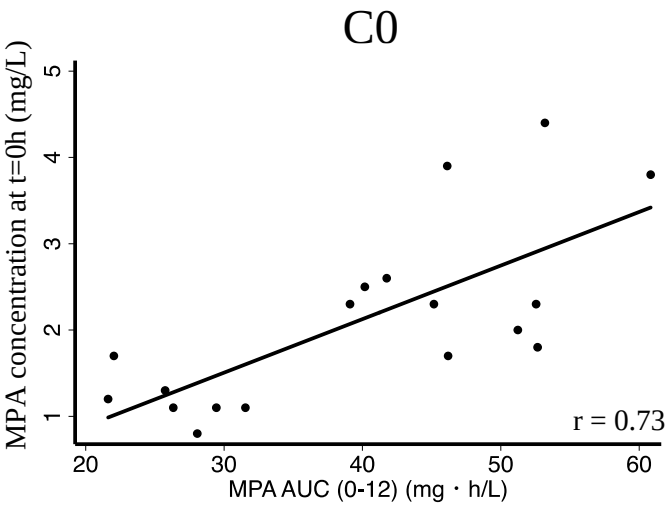

Maintenance of CR after MMF treatment
0.00
0.25
0.50
0.75
1.00
Flare cases
Initial treatment cases
Log-rank test p = 0.40
0
10
20
30
40
Observation period (months)
Number at risk
Flare = 0 9 7 7 7 0
Flare = 1 5 4 3 2 0

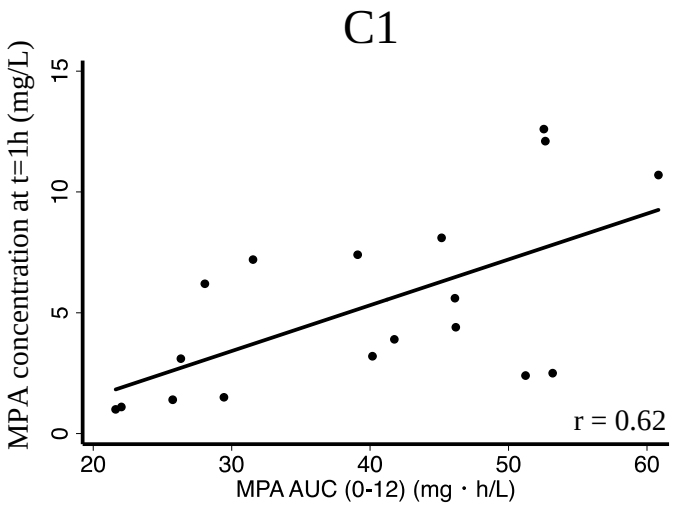
Fig. 6

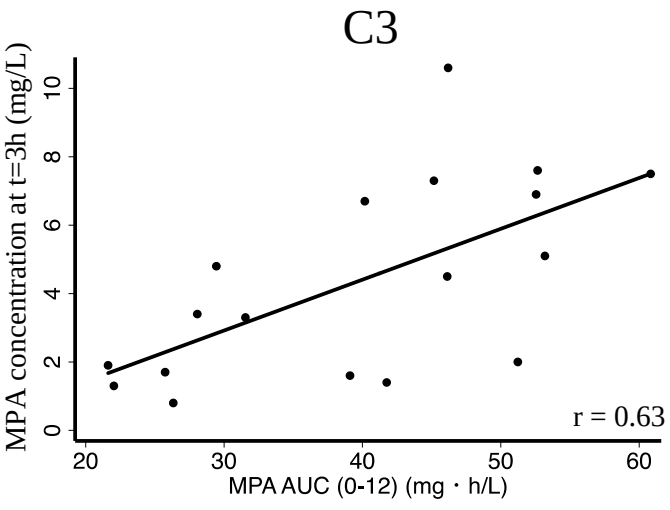
A


MPA AUC (0-12) (mg・h/L)
20
30
40
50
60
500
1000
1500
2000
MMF dose (mg/day)
r = 0.53

B


C0
MPA concentration at t=0h (mg/L)
1
2
3
4
5
20
30
40
50
60
MPA AUC (0-12) (mg・h/L)
r = 0.73


C1
MPA concentration at t=1h (mg/L)
0
5
10
15
20
30
40
50
60
MPA AUC (0-12) (mg・h/L)
r = 0.62


C3
MPA concentration at t=3h (mg/L)
0
2
4
6
8
10
20
30
40
50
60
MPA AUC (0-12) (mg・h/L)
r = 0.63

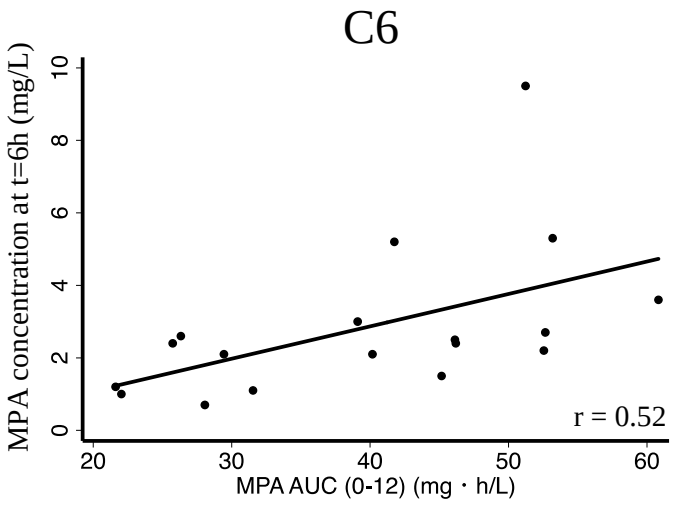
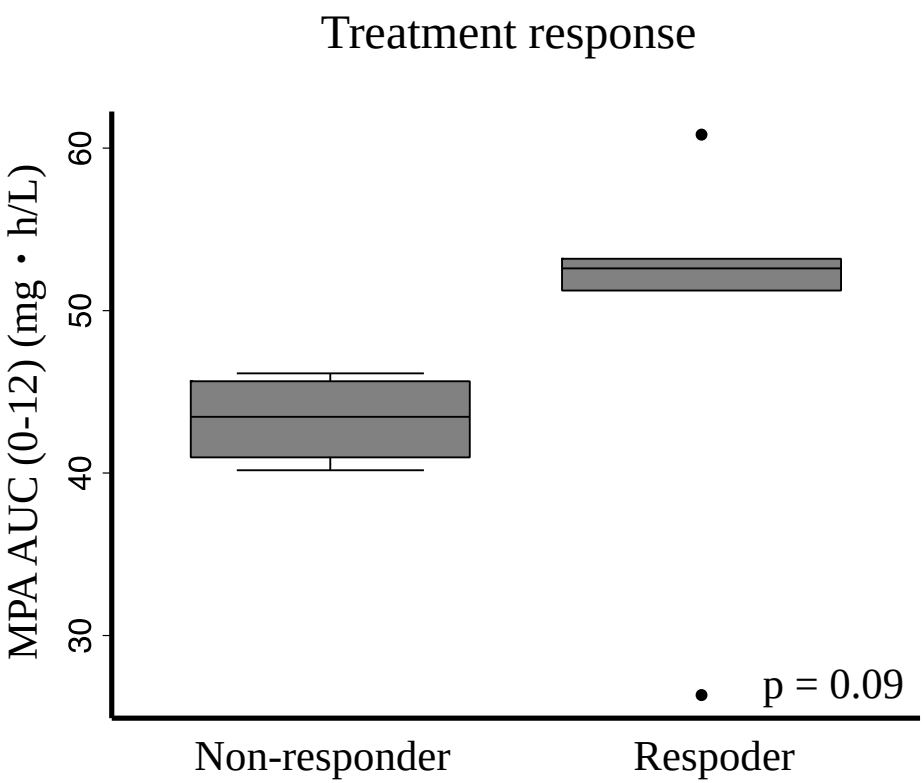
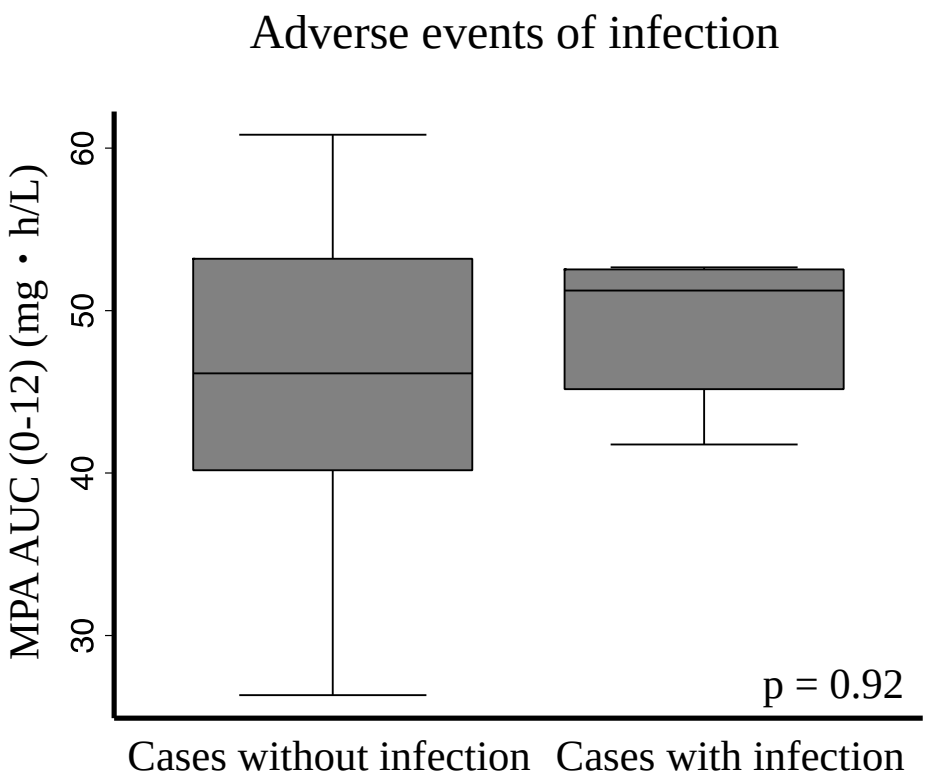

C6
MPA concentration at t=6h (mg/L)
0
2
4
6
8
10
20
30
40
50
60
MPA AUC (0-12) (mg・h/L)
r = 0.52

Fig. 7


Treatment response
MPA AUC (0-12) (mg • h/L)
30
40
50
60
p = 0.09
Non-responder
Respoder


Adverse events of infection
MPA AUC (0-12) (mg • h/L)
30
40
50
60
p = 0.92
Cases without infection
Cases with infection