Pharmacokinetics and Pharmacodynamics in Renal Transplant Recipients Under Treatment With Cyclosporine and Myfortic


ABSTRACT

Introduction. Efficacious prophylaxis of acute rejection episodes (ARE) requires adequate exposure to each component of the immunosuppressive treatment from the first days after renal transplantation. The aim of the present study was to evaluate the correlation between cyclosporine (CsA) and mycophenolic acid (MPA) exposure based upon pharmacokinetics (PK) and pharmacodynamics (PD) and 6-month biopsy-proven acute rejection (BPAR) episodes and chronic allograft nephropathy on 6 month protocol biopsies.

Patients and methods. We examined twenty-two first or second de novo renal transplant recipients treated with steroids, Sandimmune Neoral (CsA) and Myfortic (720 mg twice a day). PK (C0, C2, and AUC0–12h) for both drugs were determined on days 7, 90, and 180. Calcineurin activity, interleukin-2 and interferon-γ synthesis as well as %CEM were tested at days 7 and 180. CsA dosages were adjusted by C2 monitoring. Collected data included: BPAR during the first 6 months and Banff histological parameters on the 6-month protocol biopsies.

Results. Eighteen of 22 patients completed 1 year follow-up under treatment. The 6-month BPAR was 18% (4/22). Six-month protocol biopsies in 50% of 14 recipients showed chronic allograft nephropathy 1. At day 7, CsA C2 and AUC median values were 138 ng/mL and 6377 ng × h/mL, while C0 MPA was 1.0 μg/mL and AUC = 23.9 μg × h/mL. CsA C2 medians at 3 and 6 months were 1468 and 1720 ng/mL. MPA-AUC reached therapeutic targets at 3 months (32.3 μg × h/mL) and was 48.3 μg × h/mL at 6 months. Patients with BPAR showed lower CsA AUC (P = .06) and a significantly lower baseline inhibition of calcineurin activity (P < .005) than patients with no BPAR. An increase in mesangial matrix in 6-month protocol biopsies correlated with higher CsA C2 (P = .01). All biomarkers evaluated were significantly inhibited compared with the standard population.

Conclusions. When Myfortic is administered together with CsA, it is advisable to begin with higher doses (720 mg × 3 days) to reach adequate PK targets and improve BPAR rates. To prevent chronic allograft nephropathy, lower CsA C2 should be targeted from 3 months.

IN RENAL TRANSPLANTATION, adequate exposure to immunosuppressive drugs is critical to avoid acute rejection episodes, a risk factor for chronic rejection,1,2 and nephrotoxicity secondary to calcineurin inhibitors, the other important problem for long-term renal allograft survival.3 A multicenter, open-label, exploratory study, including clinical, pharmacokinetic, pharmacodynamic, and histological monitoring, was designed to evaluate correlations seeking to improve overall renal transplant results with 1-year follow-up.

PATIENTS AND METHODS

Study inclusion criteria were: 18 to 75-year-old first or second, living or cadaver, de novo, ABO-compatible renal transplant recipients. The immunosuppressive treatment consisted of steroids, Myfortic (720 mg twice a day), and Sandimmun Neoral adjusted for

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C2 (reference targets [ng/mL]): 1600 to 2000 for month 1, 1400 to 1600 for month 2, 1200 to 1400 for month 3, 1000 to 1200 for months 4 to 6, 800 to 1000 for months 7 to 12). The main clinical follow-up data were biopsy-proven acute rejection (BPAR) episodes and individual safety data.

Pharmacokinetic monitoring included the measurement of cyclosporine (CsA) and mycophenolic acid (MPA) trough concentrations, as well as concentrations 2 hours after drug administration and AUC0–12h (samples at 0, 30 minutes, 1, 2, 3, 4, 6, 8, 10, and 12 hours) on days 7, 90 and 180. An EMIT assay was employed for CsA concentration measurements. Plasma concentrations of MPA were analyzed by a validated and previously reported HPLC/UV method. The total run time was 12 minutes. The working range for MPA was 0.1 to 50 mg/L. Within-run and between-run imprecision ranged from 4.5% to 9.7%.

Pharmacodynamics included calcineurin activity, interleukin (IL)-2 and interferon (IFN)-γ synthesis, as well as %CEM tested at days 7 and 180. As previously described, we measured calcineurin activity in treated patients and in normal healthy control group at the morning predose and 2-hours postdose. Hypotonic lysates of PBMC were evaluated for their ability to dephosphorylate a 32P-serine-labeled 19-amino acid peptide substrate corresponding to a portion of the RII subunit of PKA. The interassay and intra-assay coefficients of variance of measurement of calcineurin activity were 8% and 5%, respectively.

IL-2 and IFN-γ production were measured by enzyme-linked immunosorbent assay in treated patients and in normal healthy control morning predose and 2-hour postdose as previously described.

Proliferation cultures with patient sera at 50% v/v final dilution were performed as previously described. In brief, spontaneously dividing CEM cells resuspended in culture medium were seeded in 96-well microtiter plates to which were added patient sera. The culture was maintained at 37°C/5% CO2 for 24 hours prior to addition of [3H]-thymidine (1.25 µCi/well) to evaluate uptake 24 hour later in a Beckman scintillation counter.

Six-month protocol biopsies performed under ultrasound guidance with a 16-gauge spring-loaded needle were processed for light microscopy. Tissue embedded in paraffin was stained with hematoxylin and eosin, periodic acid-Schiff, Masson’s trichrome, and silver methenamine. Renal lesions were blindly graded according to the 1997 Banff schema. Acute and chronic index scores were calculated as the addition of individual scores in each renal compartment. Subclinical rejection was defined as an i-score ≥1 and a t-score ≥1. Chronic allograft nephropathy was defined as a ci-score ≥1 and a ct-score ≥1.

RESULTS

Between 2003 and 2004, 22 Caucasian de novo recipients were recruited for the study including 16 men and six women of mean age 45.2 years (SD = 11.2) and body mass index mean 26.7 kg/m2 (SD = 4.6). They all received grafts from cadaveric donors who were of mean age 45.5 to 14.8 including 8/22 non-heart-beating donors. Eighteen patients completed 1-year follow-up under treatment. Four other patients included three who changed immunosuppression due to BPAR (1 of whom died) and one with delayed graft function. The 6-month BPAR rate was 18% (4/22 patients); all but one episode were steroid-sensitive. Six-month protocol biopsies performed in 14 recipients showed subclinical rejection in two patients and borderline changes in four.

Chronic allograft nephropathy I was found in 50% of cases.

Pharmacokinetics

At day 7, CsA C2 and AUC medians were 1380 ng/mL and 6377 times throughout, ng × h/mL while CsA C2 medians at 3 and 6 months were 1468 and 1720 ng/mL. C0 CsA was within target ranges at each point. C0 MPA was 1.0 µg/mL and AUC = 23.9 µg × h/mL. MPA-AUC reached therapeutic targets at 3 months (32.3 µg × h/mL) and was 48.3 µg × h/mL at 6 months. (Fig 1).

Pharmacodynamics

All biomarkers showed significant inhibition predose and at 2 hours after dosing on day 7 and 180, except for IFN-γ, which significantly recovered pre-dose at 6 months. CEM proliferation showed significant inhibition except for wide proliferation responses predose on day 7 coinciding with MPA concentrations below target levels (Fig 1).

Correlations

Patients with BPAR displayed a lower CsA AUC (P = 0.06) and a significantly lower baseline inhibition of calcineurin activity (P < 0.05) than patients with no BPAR (Fig 2). An increase in mesangial matrix in 6-month protocol biopsies correlated with higher CsA C2 (P = 0.01).
DISCUSSION

As we proposed, our study showed that adequate exposure to immunosuppressive drugs was essential in renal transplantation. We found that low C2 AUC levels at day 7 were responsible for higher activity of the target enzyme calcineurin and correlated with acute rejection episodes. A correlation of BPAR and MPA was not observed nor with IL-2 synthesis, despite low MPA levels, which may have contributed to the development of acute rejection. Our data showed that combining Sandimun Neoral with higher Myfortic doses may be needed immediately after renal transplantation to reach target levels. An initial dose of 720 mg three times a day may be advisable for the first weeks to assure correct levels, which were to be lowered before the end of the first month. These observations suggested that close pharmacokinetic and pharmacodynamic monitoring to adjust drug doses may help to avoid acute rejection episodes.

Protocol biopsies from our patients show a high prevalence of chronic allograft nephropathy at 6 months. Interestingly high C2 CsA values were observed in our patients with chronic allograft nephropathy, supporting the deleterious role of calcineurin inhibitors, which agrees with previous studies. In conclusion, combined pharmacokinetic and pharmacodynamic monitoring seems a requirement, due the fact that the equilibrium needed in renal transplantation lies in the necessity of high immunosuppression early after transplantation, together with progressive lowering of nephrotoxic drugs.

REFERENCES