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Tacrolimus is highly effective in both dual and triple therapy regimens following renal transplantation

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Abstract This open, multicenter, randomized, parallel-group study evaluated the efficacy and safety of tacrolimus-based dual and triple therapy regimens. For this 3-month study (with 12-month follow up), 491 adult renal transplant patients were randomized and received either dual therapy (tacrolimus/corticosteroids; 246 patients) or triple therapy (tacrolimus/corticosteroids/azathioprine; 245 patients). Patient survival rates at months 3 and 12 were 99.2 (dual) vs 99.6% (triple) and 97.8 vs 98.7%, respectively. Graft survival rates at months 3 and 12 were 94.1 (dual) vs 95.4% (triple) and 92.8 vs 93.3%, respectively. After 3 months, the incidences of treated acute rejection were 28.8 (dual) and 29.7% (triple); and 7.6 (dual) and 5.4% (triple) for corticosteroid-resistant acute rejections. Between months 4 and 12, three new first rejections were reported, (dual: 2, triple: 1). For leukopenia (1.3 vs 11.7%; $P < 0.001$) and anemia (14.8 vs 23.0%, $P = 0.026$), significantly higher incidences were reported in the triple therapy group. The incidence of de novo insulin-dependent diabetes was 5.6 (dual) and 4.0% (triple) at month 3. In terms of efficacy, no difference between the treatment groups was observed.

Key words Tacrolimus · Azathioprine · Kidney transplantation · Dual vs triple therapy · Rejection

Introduction

Aiming to reduce the incidence of rejection after renal transplantation, a current trend emphasizes the use of multiple drug regimens. Combinations of "cornerstone" immunosuppressants, such as tacrolimus or cyclosporine and corticosteroids, are supplemented with adjunctives, such as azathioprine, mycophenolate mofetil, sirolimus, or antibody preparations. In large multicenter clinical trials comparing tacrolimus-based therapies with standard cyclosporine based therapies, substantially lower rates of acute rejection were observed with tacrolimus treatment [3, 4]. In these studies, a triple drug regimen consisting of tacrolimus, corticosteroids, and azathioprine was used. A recent single-center study in the USA investigated the clinical course of 395 renal allograft recipients treated with tacrolimus and corticosteroids with or without azathioprine [5, 6]. The authors report no significant difference between the treatment groups in respect to both efficacy and safety. However, the study suffered from a high crossover rate between treatment groups; in the dual therapy group 17% of patients received azathioprine, and 40% of patients on triple therapy discontinued azathioprine administration [6]. The present study aimed to evaluate the efficacy and safety of immunosuppressive regimens based on tacrolimus and tapered corticosteroids with or without azathioprine in a European setting.

Patients and methods

Study design

Eleven centers in Italy and 25 centers in Spain participated in this open, randomized, phase III, parallel-group study conducted between October 1996 (first patient in) and April 1998 (final visit date). Patients were included in this study if they were over 18 years of age, had end-stage renal disease, and were suitable for primary renal transplantation or retransplantation with a cadaveric kidney graft. Excluded from the study were patients who were pregnant, allergic, or intolerant to antimetabolites, HCO-60 or structurally related compounds, steroids, macrolide antibiotics or tacrolimus, if they were HIV, HBV, or HCV positive, or received an ABO-incompatible graft. Patients were randomly assigned in a 1:1 ratio to receive either a tacrolimus-based dual (tacrolimus/corticosteroids) or triple (tacrolimus/corticosteroid/azathioprine) immunosuppressive regimen. Randomization was preoperative. The study was conducted in accordance with the Declaration of Helsinki. Approval was obtained from local ethics committees and informed consent was provided by each patient prior to enrolment.

Treatment protocol

The initial oral tacrolimus dose was 0.2 mg/kg per day; the dose was subsequently adjusted to maintain a target whole blood trough level of 8–15 ng/ml. Methylprednisolone was given on day 0 as a 500 mg i.v. bolus and on day 1 as a 125 mg i.v. bolus. Oral prednisone was tapered from 20 mg/day at day 2 to 5 mg/day at day 43. In the triple group, azathioprine was administered at day 0 as a 2 mg/kg i.v. bolus and from day 1 until study end, 1–2 mg/kg was administered orally once daily. Tacrolimus blood level was monitored by a microparticle enzyme immunoassay (IMx).

Table 1 Demographic and baseline characteristics

	Dual therapy ($n = 236$)	Triple therapy ($n = 239$)
Age (median, years) (range)	46.0 (21–69)	45.0 (19–68)
Male/female	153/83	154/85
Hypertension	169 (71.6%)	177 (74.1%)
Mean HLA-antigen mismatches A/B/DR	1.11/1.26/0.68	1.11/1.21/0.74
Mean cold ischemia time (h, range)	18.0 (4–40)	17.6 (6–36)
PRA grade		
0–< 50%	230 (99.1%)	234 (99.2%)
50–100%	2 (0.9%)	2 (0.8%)
Not recorded	4	3

Study end points

The efficacy end points were patient and graft survival, incidence of first acute rejection, and incidence of first corticosteroid-resistant acute rejection. Safety was assessed by monitoring adverse events, laboratory parameters, and vital signs. Graft failure was defined as the need to return to dialysis, nephrectomy, or death.

Statistical evaluation

The sample size was based on an assumed incidence of first acute rejections in 25% of renal graft recipients on a triple therapy within 3 months after transplantation. It was estimated that, by using a 1:1 randomization, a total of 400 evaluable patients should give this study a power of at least 80% to detect a difference of 13.5% in the incidence of first acute rejection using a two-sided significance test (alpha = 0.05). The incidence rates (rejection episodes, adverse events) in the treatment groups were compared using the chi-squared test or Fischer's exact test.

Results

Four hundred and ninety-one patients were recruited for this study and, of these, 16 patients never received the study drug or were not transplanted. Thus, the intent-to-treat cohort comprised 475 patients, 236 patients on dual therapy and 239 patients on triple therapy. The treatment groups were similar with respect to baseline demographic characteristics (Table 1). The frequencies of end-stage renal diseases were similar in both treatment groups (Table 2). The 3-month observation period was completed by 211/236 (89.4%) of patients in the

Table 2 Cause of end-stage renal disease

	Dual therapy ($n = 236$)	Triple therapy ($n = 239$)
Chronic glomerulonephritis	74 (31.4%)	77 (32.2%)
Interstitial pyelonephritis	30 (12.7%)	27 (11.3%)
Polycystic disease	28 (11.9%)	30 (12.6%)
Nephrosclerosis	15 (6.4%)	13 (5.4%)
Diabetes mellitus type I and II	15 (6.4%)	9 (3.8%)
Analgesic nephropathy	1 (0.4%)	2 (0.8%)
Other/unknown	73 (30.9%)	81 (33.9%)

Table 3 Incidence of rejection (based on patients)

	Triple therapy, 3-months ($n = 236$)	Dual therapy, 3-months ($n = 239$)
Treated acute rejection	68 (28.8%)	71 (29.7%)
Corticosteroid-resistant acute rejection	18 (7.6%)	13 (5.4%)
Antibody-sensitive acute rejection	8 (3.4%)	8 (3.3%)
Refractory acute rejection[a]	11 (4.7%)	5 (2.1%)
Treated biopsy-confirmed acute rejection	39 (16.5%)	37 (15.5%)

[a] Rejections ongoing at the end of months 3

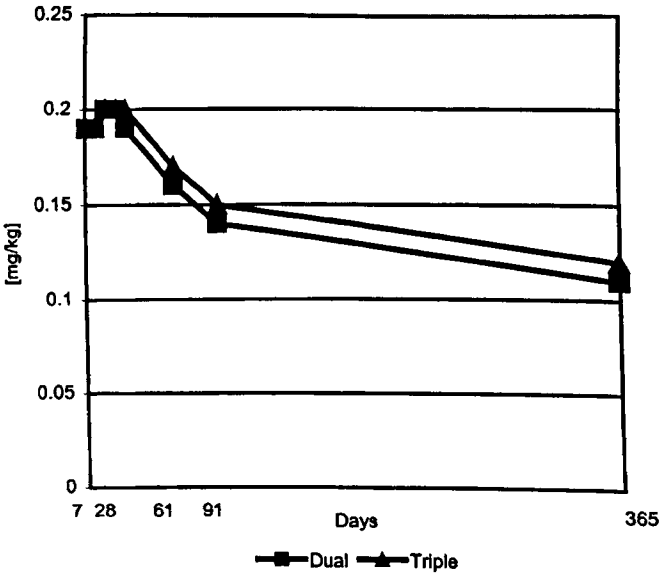

Fig. 1 The mean daily tacrolimus dose decreased during the course of the study from an initial 0.19 mg/kg for both study groups during the 1st postoperative week to 0.11 (dual) and 0.12 (triple) at study end

dual therapy group and 212/239 (88.7%) of patients in the triple therapy group. The 12-month follow up was completed by 207/236 (87.7%) of patients in the dual and 209/239 (87.4%) of patients in the triple therapy group. At the time of the 12-month follow up, 5 patients of the dual therapy group received azathioprine, whereas azathioprine was discontinued in 56 patients of the triple therapy group.

Efficacy

At 3 months, patient survival and graft survival rates were 99.2 and 94.1 in the dual therapy and 99.6 and 95.4% in the triple therapy group, respectively. During the 12-month observation period, five patients in the dual therapy group and three patients in the triple therapy group died. After 12 months, patient survival and graft survival rates were 97.8 and 92.8% (dual) vs 98.7 and 93.3% (triple), respectively. Until the end of month 3, 68/236 (28.8%) of patients from the dual group and 71/239 (29.7%) of patients in the triple group were treated for acute rejection (Table 3). Corticosteroid-resistant acute rejections were reported by 7.6 and 5.4% of patients in the dual and triple groups, respectively. The treatment groups did not differ with respect to the severity of rejection episodes (data not shown). Between months 4 and 12, new first acute rejections were experienced by two patients in the dual therapy group and one patient in the triple therapy group. Mean serum creatinine concentrations at day 91 were 155 (dual) and 152 μmol (triple), and 143 (dual) and 135 μmol (triple) at month 12. Mean total cholesterol levels remained at screening values (dual: 5.23 ± 1.24 mmol/l; triple: 5.20 ± 1.31 mmol/l) throughout the observation period. At month 3 and month 12, total cholesterol levels (± SD) were 5.20 ± 1.07 and 5.47 ± 1.25 mmol/l in the dual therapy group compared with 5.10 ± 1.12 and 5.34 ± 1.00 mmol/l in the triple therapy group. Lipid-lowering drugs were administered to 5/211 (2.4%) of patients in the dual therapy group and to 4/212 (1.9%) of patients in the triple therapy group at month 3. After 12 months, 18/207 (11.5%) of patients in the dual therapy group and 18/209 (11.6%) of patients in the triple therapy group received antihyperlipidemic medication.

Dosing and blood levels

At day 1, the mean daily oral tacrolimus dose was 0.19 mg/kg for both treatment groups. At day 91, the dose was reduced to 0.14 mg/kg in the dual therapy group and 0.15 mg/kg in the triple therapy group. At month 12, the mean daily oral tacrolimus dose was 0.11 (dual) and 0.12 mg/kg (triple; Fig. 1). Mean tacrolimus blood levels (± SD) at day 1 were 15.4 ± 9.6 and 16.3 ± 10.4 ng/ml for the dual and triple groups (Fig. 2). At day 91, the corresponding tacrolimus whole blood trough levels (± SD) were 12.0 ± 3.9 ng/ml in the dual therapy group and 12.4 ± 4.0 ng/ml in the triple therapy

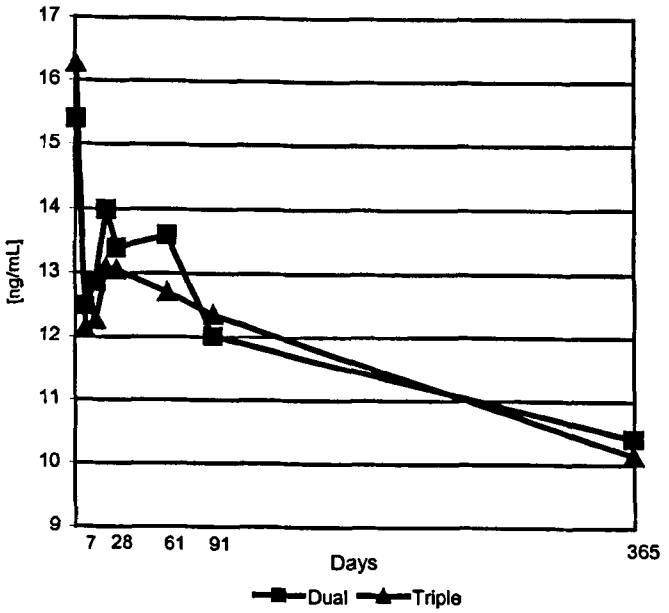

Fig. 2 The mean tacrolimus whole blood trough levels decreased from 12.49 (dual) and 12.15 ng/ml (triple) at day 1 to 10.41 (dual) and 10.14 ng/ml (triple) at the end of the study

group. At month 12, the mean tacrolimus whole blood levels were at 10.4 ± 3.33 (dual) and 10.1 ± 3.18 ng/ml (triple).

Adverse events

The most frequently reported adverse events in both treatment groups were infection (dual: 109/236 (46.2%) vs triple: 110/239 (46.0%); P = ns), hypertension (67/236 (28.4%) vs 57/239 (23.4%); P = ns), urinary tract infection (64/236 (27.1%) vs 58/239 (24.3%); P = ns), and tremor (40/236 (16.9%) vs 43/239 (18.0%); P = ns). Leukopenia (3/236 (1.3%) vs 28/239 (11.7%); $P < 0.001$) and anemia (35/236 (14.8%) vs. 55/239 (23.0%); $P = 0.026$) were reported significantly more often in the triple therapy group. At month 3, insulin for de novo posttransplant diabetes mellitus was required by 11/195 (5.6%) and 8/200 (4.0%) of patients in the dual and triple groups.

Discussion

In a previous single-center study of similar design, one of the confounding factors was the high number of patients who switched treatment arms during the study [5, 6]. The present study succeeded in keeping the number of treatment switches to a minimum. Only 5 patients of the dual therapy group had azathioprine added to their immunosuppressive regimen, whilst azathioprine was discontinued in 56 patients of the triple therapy group.

The treatment groups exhibited similar baseline characteristics. Both treatments resulted in high patient and graft survival after both 3 and 12 months. Low acute rejection rates after both 3 and 12 months were observed in both treatment groups. The incidence of biopsy-proven rejections was low during the observation period of 12 months.

With respect to safety, both treatments showed a similar profile. After month 3, the only differences between the treatment groups were found for leukopenia and anemia. Both adverse events were more common in the triple therapy group than in the dual therapy group. This result can be attributed to the mode of action of azathioprine [1, 7]. We therefore conclude, that in patients receiving tacrolimus-based triple therapy who develop either leukopenia or anemia, azathioprine can safely be discontinued.

The incidence of new onset insulin-dependent diabetes mellitus after 3 months was low (dual therapy 5.6%, triple therapy 4.0%). As reported previously, tacrolimus is neutral in respect to posttransplant cholesterol and triglyceride levels [2]. This finding is supported by the present study since only a small number of patients received lipid-lowering drugs after 3 and 12 months of tacrolimus-based immunosuppressive therapy.

Our study showed that tacrolimus-based dual and triple therapy regimens are efficacious and safe. The addition of azathioprine to a dual therapy of tacrolimus and corticosteroids did not increase the efficacy in terms of the prevention of acute rejection.

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References

1. Bergan S (1998) Azathioprine monitoring in renal transplantation, University of Oslo, Rikshospitalet. ISBN 82-7633-106-8
2. Claesson K, Mayer AD, Squifflet JP, et al (1998) Lipoprotein patterns in renal transplant patients: a comparison between FK 506 and cyclosporine A patients. Transplant Proc 30: 1292–1294
3. Mayer AD, Dmitrewski J, Squifflet JP, et al (1997) Multicenter randomized trial comparing tacrolimus (FK506) and cyclosporine in the prevention of renal allograft rejection. Transplantation 64: 436–443
4. Pirsch JD, Miller J, Deierhoi MH, et al (1997) A comparison of tacrolimus (FK506) and cyclosporine for immunosuppression after cadaveric renal transplantation. Transplantation 63: 977–983
5. Shapiro R, Jordan ML, Scantlebury VP, et al (1995) A prospective randomized trial of FK506-based immunosuppression after renal transplantation. Transplantation 59: 485–490
6. Shapiro R, Jordan ML, Scantlebury VP, et al (1995) A prospective, randomized trial of FK506/prednisone vs FK506/azathioprine/prednisone in renal transplant patients. Transplant Proc 27: 814–817
7. Walker RG (1994) Nonspecific immunosuppression: azathioprine and steroids. In: Morgan JP (ed) Kidney transplantation. Principles and practice, 4th edn. Saunders, pp 202–214