transplants 1992. Los Angeles: UCLA Tissue Typing Laboratory, 1993: 237.
14. Scornik JC, Brunson ME, Schaub B, et al. The crossmatch in renal transplantation: evaluation of flow cytometry as a replacement for standard cytotoxicity. Transplantation 1994; 57: 621.
15. Mahoney RJ, Norman DJ, Colombe BW, et al. Identification of high- and low-risk second kidney grafts. Transplantation 1996; 61: 1349.
16. Scornik JC, Bray RA, Pollack MS, et al. Multicenter evaluation of the flow cytometry T-cell crossmatch. Transplantation 1997; 63: 1440.

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REASONS FOR LONG-TERM USE OF STEROID IN PRIMARY ADULT LIVER TRANSPLANTATION UNDER TACROLIMUS[1,2]

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Background. **Tacrolimus is a potent immunosuppressive agent that provides higher freedom from acute and chronic rejection than cyclosporine after liver transplantation (LTx). Initially, a steroid-free state was observed in about 70% of patients at 1 year; this did not change over the next 5 years. The present study identifies the various reasons why the remaining 30% of adult patients still require steroids even after 5 years after successful LTx.**

Method. **Eight hundred thirty-four consecutive patients who underwent LTx between August 1989 and December 1992 were included in this study. Four hundred ninety-nine patients were alive in January 1999 and were available for this study. The dose of steroid and the reason for steroid use were retrospectively determined from the clinical records.**

Results. **Three hundred sixty-five patients (73.1%) were off steroid, whereas 134 patients (26.9%) were receiving prednisone (mean dose was 6.4±3.7 mg/day) at the time of the study. Four hundred and eight-four patients (97%) were off prednisone at some time after LTx; however, in 119 (23.8%) patients, steroids were reintroduced. Fifteen patients (3%) continued to receive prednisone; eight receive prednisone due to reluctance of the local physician to withdraw the medication; in five patients, the prednisone was not withdrawn because these patients were on cyclosporine; in the remaining two patients, repeated attempts to withdraw steroid resulted in a rise in liver function test. In the 49 (36.6%) of 119 patients in whom the steroid was reintroduced, it was restarted secondary to pathologically proven or clinically suspected rejection (group I). In five patients steroid was reintroduced for abnormal liver function after being off immunosuppression for treatment of a posttransplantation lymphoproliferative disorder. Six patients were noncompliant with their immunosuppressive medication, and the steroid was reintroduced to control rejection. Steroids were reintroduced in 30 patients (22.4%) for recurrence of original disease: primary biliary cirrhosis (n=19), sclerosing cholangitis (n=6), and autoimmune hepatitis (n=5) (group II). In 24 patients (20.2%), steroids were reintroduced to lower the dose of tacrolimus secondary to nephrotoxicity. Six of these patients received kidney transplantation (group III). In 16 patients (13.4%) the steroid was reintroduced for concomitant medical problems, consisting of ulcerative/Crohn's colitis (n=6), adrenal insufficiency (n=5), hematological disorders (n=3), dermatitis (n=1), and rheumatoid arthritis (n=1) (group IV).**

Conclusion. **Ninety-seven percent of patients under tacrolimus were weaned off steroid; however, 23.8% required steroid reintroduction for late rejection, recurrence of autoimmune process(es), renal impairment, or the concomitant presence of other medical conditions. Although the use of other immunosuppressive agents may reduce the rate of reintroduction of steroid, long-term sustained freedom from steroid may**

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not be possible in all patients under tacrolimus secondary to these conditions.

The beneficial effect of tacrolimus in liver transplantation (LTx) is well recognized, particularly in terms of the overall reduction of episodes of acute rejection, steroid-resistant rejection, and refractory rejection (*1–3*). In our preliminary observation of 1,000 consecutive primary LTx patients reported in 1995, freedom from steroid was observed in 70% of patients 1–4 years post-LTx (*4*). On subsequent examination of the same population 3 years later, there was no further increase in freedom from steroid, an interesting observation given that we have an active immunosuppression withdrawal protocol at our institution for patients with stable allograft function beyond 5 years (5-7).

Steroids are known to cause many metabolic changes including hypertension, diabetes, obesity, osteoporosis, and hypercholesteremia (*8–10*). The aim of the present study is to examine the rate of freedom from steroid under tacrolimus, the dose of steroid use, and to identify the reasons for use of steroid in adults (age>18 years) 5 years or more after primary LTx.

PATIENTS AND METHODS

Between August 1989 and December 1992, 834 adults (age >18 years) underwent primary LTx under tacrolimus. All patients were followed until January 1999, with a mean follow-up of 7.6±0.86 years (range 6 to 9.3). Current immunosuppression dosing of all living patients was reviewed. The study identified the dose and reason for continued steroid use for those still receiving it. The tacrolimus/steroid protocols used at our institution have been previously described elsewhere (*3*, *4*, *11*, *12*).

RESULTS

Three hundred thirty-five patients (40.2%) died during the mean follow-up of 7.6 years, leaving 499 patients available for study. There were 276 (55.3%) males and 223 (44.7%) females. The mean age was 49±12 years at the time of transplantation and 56±12 years at the time of evaluation. The primary diagnosis of the patients living at the time of the study is shown in Table 1.

Three hundred sixty-five patients (73.1%) are free from steroid; 134 (26.9%) were still on steroid and were examined in further detail. The dose of prednisone was ≤5 mg/day in 93 (69.4%), between 6 and 10 mg/day in 32 patients (23.9%), and >10 mg/day in only 9 (6.7%). The mean daily prednisone dose is 6.4±3.7 mg (median 5 mg: range 1.25–20). Of the 134 patients on steroid at the time of this study, all but 15 patients (11.3%) were off prednisone at some time after LTx. In the remaining 119 patients, 16 different reasons were identified for reintroduction of drug and were divided into 5 different groups (summarized in Table 2).

Reasons for not withdrawing steroid. In 15 patients prednisone was never withdrawn after LTx. Eight patients were managed by local physicians—several outside this country[em]and our center had inadequate follow-up. Prednisone was not withdrawn in five patients because they were on cyclosporine. In the remaining two patients, repeated attempts at lowering the steroid led to an increase in hepatic enzyme activity; hence prednisone was continued (group I).

Reasons for reintroduction of steroid. Late rejection: In 49 (36.6%) of 134 patients, prednisone was reinstituted for either biopsy proven (n=24; 49%) rejection or a rise in hepatic enzymes, i.e., clinically suspected mild rejection (n=25; 51%) (group II). The mean dose of prednisone in this group was 6.3±3.9 mg/day (median 5; range 1.25 to 20). In six patients the cause of late rejection was considered to be documented noncompliance with the immunosuppressive medications. In another five patients the immunosuppression was either discontinued or was markedly reduced to control posttransplant lymphoproliferative disorder that subsequently led to clinical

TABLE 1. Primary diagnosis

Indicatrions	n	%
PNCE	111	22.2
HCV	82	16.4
PBC	62	12.4
Cryptogenic	56	11.2
PSC	44	8.8
HBV	41	8.2
Hepatic malignancy	30	6
FHF	19	3.8
Autoimmune	17	3.4
Secondary biliary cirrhosis	12	2.4
Budd chiari	7	1.4
A-I-A deficiency	7	1.4
Other (Wilsons-3, hemochromotosis-3, biliary atresia-3, trauma-1, unknown-1)	11	2.2
Total	499	

A-1-A, alpha 1 antitrypsin; FHF, fulminant hepatic failure; HBV, hepatitis B virus; HCV, hepatitis C virus; PNCE, ethanol induced post necrotic cirrhosis.

TABLE 2. Reasons for use of steroid

Group	Reason	n	%
I	Steroid not withdrawn	15	11.9
	Poor follow-up long distance	8	
	Cyclosporine	5	
	Inability due to increase in liver function	2	
II	Late rejection (reinstitution of steroid)	49	36.57
	Noncompliance	6	
	Posttransplant lymphoproliferative disorder	5	
	Unknown reason for late rejection	38	
III	Recurrence of primary disease (reinstitution of steroid)	30	22.39
	Primary biliary cirrhosis	19	
	Primary sclerosing cholangitis	6	
	Autoimmune disease	5	
IV	Renal impairment (reinstitution of steroid)	24	17.91
	Ongoing renal impairment	18	
	Kidney transplantation	6	
V	Concomitant medical conditions (reinstitution of steroid)	16	11.94
	Colitis	5	
	Adrenal insufficiency	5	
	Hematological disorder	3	
	Rheumatoid arthritis	1	
	Dermatitis	1	
Total		134	

rejection and reintroduction of steroid. In the remaining patients the cause of late rejection was not determined. Five patients in this group were also on azathioprine (25 to 75 mg/day), and two others were on mycophenolate mofetil (MMF) (1 g/day). Nearly all patients normalized the biochemical abnormality in the liver function tests.

Recurrence of disease: In 30 patients (22.4%) steroid was reintroduced for biopsy-proven disease recurrence, consisting of primary biliary cirrhosis (PBC) (n=19; 14.1%), primary sclerosing cholangitis (PSC) (n=6; 4.5%), and autoimmune hepatitis (AI) (n=5; 3.7%) (group III). The mean dose of steroid was 6.8±3.4 mg/day (median 5, range 5 to 20). Four patients in this group were also on azathioprine (50 to 75 mg/day), and another four were on MMF (500 to 1500 mg/day). Overall freedom from steroid for PBC in this study population was 40 (64.5%) of 62; that for PSC was 31 (70.5%) of 44 and for AI was 8 (47%) of 17.

Renal impairment and kidney transplantation: Twenty-four (17.9%) of the patients were on prednisone for renal impairment (n=18; 13.4%) to allow a lower concomitant dose of tacrolimus; the remaining six (4.5%) received kidney transplants (group IV). Three patients with renal impairment are also on azathioprine (50 to 100 mg/day), and one patient is on MMF (1000 mg/day). The mean dose of prednisone in this group was 5.3±1.9 mg/day (median 5, range 2.5 to 10).

Other concomitant medical conditions: Sixteen (11.9%) of the patients had a concomitant medical condition that required the reintroduction of prednisone (group V). This consisted of colitis (ulcerative=5, Crohn's=1; 4.5%), adrenal insufficiency (n=5; 3.0%), hematological disorder (hemolysis=2, idiopathic thrombocytopenic purpura=1; 2.2%), dermatitis (n=1; 0.7%), and rheumatoid arthritis (n=1; 0.7%). The mean dose of steroid for this group was 7.6 + 5.1 mg/day (median 5, range 2.5 to 20).

Rate of hypertension, diabetes, and hyperlipidemia. The rate of hypertension was 45.2% in patients who were off steroid and 50.7% for the patients who were on steroid; the difference was not significant. There was a slight increase in the rate of hypertension with increasing dose of steroid, but it was not statistically significant (Table 3). Similarly the rate of insulin-dependant diabetes mellitus was 15% in patients who were off steroid and 17.9% for patients who were on prednisone. There was no trend for the incidence of diabetes correlated with the dose of steroid in this study.

Cholesterol were measured only in 138 (27.7%) patients, 26 (19.2%) in the steroid group and 112 (30.7%) in the nonsteroid group. Also triglyceride levels were available in only 113 (22.6%) patients, 21 (15.7%) in the steroid group and 92 (25.2%) in the nonsteroid group. There was no significant difference in this study in levels of cholesterol or triglycerides, but information was available in only a limited number of patients (Table 3).

Concomitant use of other immunosuppressive agents. Concomitant use of azathioprine and MMF for various groups of patients on steroid is shown in Table 4. Fifteen patients were on azathioprine, and six patients were on MMF without steroids. The mean dose of azathioprine was 53.5±24 mg/day [100 mg/day (n=2), 75 mg/day (n=2), 50 mg/day (n=7), and 25 mg/day (n=4)]. The mean dose of MMF was 1000±500 mg/day [2000 mg/day (n=1), 1000 mg/day (n=3), and 500 mg/day (n=2)]. Three hundred and forty-four patients (68.9%) were on either cyclosporine or tacrolimus monotherapy.

Biochemical parameter. There was no difference in biochemical parameters indicative of liver function (mean serum bilirubin, aspartate aminotransferase, alanine aminotransferase, alkaline phosphatase, and gamma-glutamyl transferase) for both groups of patients who were on steroid or without steroid (Table 5). Similarly mean blood urea nitrogen and mean serum creatinine was also comparable at the last follow-up for both groups as shown in Table 5.

DISCUSSION

At our institution we do not have any fixed deliberate policy for a definitive time after LTx for withdrawing steroid. Instead, patients are managed on an individual basis depending on their liver function and previous history of rejection. Since the metabolic, cosmetic, and noncosmetic effects of steroid are well recognized, it is the first drug that is usually weaned under optimal conditions, the steroid weaning process starts 2 weeks after LTx. If there is no evidence of rejection within 2 weeks and the patient's liver function tests are nearly normal, prednisone is reduced from 20 mg/day (starting baseline dose) to 15 mg/day. This process continues for the rest of the follow-up. By 3 months after transplantation about 50% of patients are off steroid; by 1 year it is about 70%. However, because of clinical development, the steroid dose must be increased or restarted in some.

Steroid withdrawal after LTx has been shown to be of benefit, resulting in less hypertension, diabetes, obesity, and elevated cholesterol levels (*8–10*). Tisone et al. (*13*) in a prospective randomized study of 20 patients under cyclosporine and azathioprine with or without steroid showed no difference in the rate of rejection or survival and found better liver and renal function without steroid. However the comparison was made with only a 20-mg of prednisone per day dose of steroid. In our early trial when we compared high-dose steroid induction versus low-dose steroid (20 mg/day vs. 1000 mg induction and 600 mg taper over the next 5 days)

TABLE 3. Hypertension, diabetes, and hyperlipidemia in relation to prednisone dose

Prednisone dose	Hypertension	Diabetes	Cholesterol[a]	Triglyceride[b]
0.00 (n=365)	165 (45.2%)	55 (15%)	178±45	163.2±121
>0 ≤5 (n=93)	45 (48.4%)	17 (18%)	182±40	151.9±72.6
>5 ≤10 (n=32)	17 (53.1%)	6 (18.8%)	186±54	108.8±53.2
>10 (n=9)	6 (66.7%)	1 (11.1%)	NA	NA
All Steroid (n = 134)	68 (50.7%)	24 (17.9%)	180±51	137.1±75.1

[a] Available 26 (19.4) in steroid group and 112 (30.7%) in nonsteroid groups.
[b] Available in 21 (15.7%) steroid group and 92 (25.2%) nonsteroid group.
NA, not available.

TABLE 4. Mean dose of prednisone and use of azathioprine and MMF

Group	Mean prednisone dose mg/day	Azathioprine, n (dose mg/day)	MMF n (dose mg/day)
Group I	5.3+1.9	0	0
Group II	6.3+3.9	5 (25, 50, 50, 75, 75)	2 (1000, 1000)
Group III	6.8+3.4	4 (50, 50, 50, 75)	4 (500, 500, 1000, 1500)
Group IV	5.3+1.9	3 (50, 50, 100)	1 (1000)
Group V	7.6+5.2	0	1 (500)

TABLE 5. Liver and renal function

	On steroid (Mean±SD)	Off steroid (Mean±SD)
Liver function test		
Bilirubin mg/dl	0.6±0.8	.7±.3
AST IU/L	33.8±28.4	31±18
ALT IU/L	36±27	37±32
ALKP IU/L	82.6±82.3	101±78
GGT IU/L	76.3±96.7	94±14.3
Renal function		
Blood urea mg/dl	27.4±14	24±12
Creat mg/dl	1.8±1.3	1.6±1.3

ALKP, alkaline phosphatase; ALT, alanine amino transferase; AST, aspartate amino transferase; GGT, gamma-glutamyl transferase.

there was a significant reduction in rejection rate; however morbidity was not compared in the study (*11*). Gomez et al. (*9*) showed successful steroid withdrawal in 86 selected patients 1 year after LTx with stable liver function and found improvement in hypertension, cholesterol, and bone mineral density in the lumber spine under cyclosporine without the need for azathioprine. Padbury et al. (*14*) from U.K. reported successful withdrawal of steroid in 85% of patients 3 months after LTx with improvement in hypertension and rate of infection. Similar observations were made by Punch et al. (*15*) with 88% successful steroid withdrawal at mean a follow-up of 13.8 months. They observed a decrease in hypertension, cholesterol, and weight gain. Ramirez et al. (*16*) have demonstrated a 90% success rate of withdrawal of steroid at 6 months after LTx under cyclosporine and tacrolimus.

In the present study there were no protocols involved to wean the steroid by introducing azathioprine, MMF. or the recently approved drug rapamycin. It is quite possible that more patients could be weaned off by adding any of the above agents. McDiarmid et al. (*17*) have successfully withdrawn steroid in selected post-LTx patients on cyclosporine by adding azathioprine. Similarly Stegall et al. (*8*) reported the withdraw of steroid 14 days after LTx by using MMF in conjunction with either tacrolimus or cyclosporine (or Neoral) and found beneficial effects in terms of decreased rate of diabetes and a lower rate of cholesterol and hypertension with no increased immunological risk to the allograft. DeCarlis et al. (*18*) used a quadruple drug regimen: rabbit antithymocyte globulin and steroid induction with cyclosporine and azathioprine. At 3 months patients were randomized to complete steroid withdrawal or long-term low-dose steroid. The authors found a significant benefit in terms of hypertension, diabetes, bone complications, and lower cholesterol level for the group of patients off steroid. However, it is important to realize that these agents that are being substituted for steroid are not free from side effects. The metabolic, cosmetic, and noncosmetic benefits of steroid avoidance, particularly for the majority of patients who are on 5 mg or less per day, need to be weighed against the potential side effects of azathioprine, MMF, and rapamycin. An extremely careful, closely monitored prospective study will need to be conducted to determine the real benefit for these patients, including the questions of the side effects of these other immunosuppressive medications. However, if the patient has existing problems with osteoporosis, obesity and mild hypertension, and/or mild diabetes mellitus, he/she may benefit from maximal steroid reduction. However, the long-term goal has to be monotherapy or an immunosuppression-free state rather than polypharmacy therapy. An immunosuppression-free state seems to be possible only in a small percentage of patients (*19*). However, hopefully LTx with the simultaneous infusion of donor bone marrow to promote better microchimerism will prove to be useful in allowing an immunosuppression-free state; until then monotherapy seems to be the only other best option (*20*). McMaster et al. (*21*) has summarized the changing goals in immunosuppression with tacrolimus and cyclosporine.

MMF and rapamycin are approved by the FDA for use in kidney transplantation with cyclosporine or Neoral only. Its safety with tacrolimus in liver transplantation has not yet been conclusively evaluated, and many third-party payers are not willing to reimburse the cost. In any event, these approaches of combining tacrolimus with MMF and rapamycin in liver transplant patients have scientific basis and offer potential advantages in allowing the weaning of steroid and may prove to provide other advantages as well, particularly with regards to obesity, hypertension, hyperglycemia, and hypercholesteremia.

In conclusion, 73% of patients were off steroid in the present series; 18.1% were on prednisone ≤5 mg/day, 6.4% were on prednisone between >5 and ≤10 mg/day, and 1.8% of patients were on prednisone >10 mg/day. Ninety-seven percent of patients were completely off prednisone at some time after LTx. However, up to 24% of the patients may need reinstitution of small doses of steroids for the various reasons outlined above but may be minimized by the concomitant use of the newly approved immunosuppressive agents with tacrolimus. It seems that compliance issues, late rejection, management of a recurrent autoimmune process, and associated other medical conditions will continue to cause problems for the successful complete and sustained long-term freedom from steroids in a small percentage of the cases.

REFERENCES

1. Anonymous. A comparison of tacrolimus (FK 506) and cyclosporine for immunosuppression in liver transplantation. The U.S. Multicenter FK506 Liver Study Group [see comments]. N Engl J Med 1994; 331(17): 1110.
2. Anonymous. Randomised trial comparing tacrolimus (FK506) and cyclosporin in prevention of liver allograft rejection: European FK506 Multicentre Liver Study Group. Lancet 1994; 344(8920): 423.
3. Fung JJ, Eliasziw M, Todo S, et al. The Pittsburgh randomized trial of tacrolimus compared to cyclosporine for hepatic transplantation. J Am Coll Surg 1996; 183(2): 117.
4. Jain AB, Fung JJ, Todo S, et al. One thousand consecutive primary orthotopic liver transplants under FK 506: survival and adverse events. Transplant Proc 1995; 27(1): 1099.

5. Jain A, Reyes J, Kashyap R, et al. Liver transplantation under tacrolimus in infants, children, adults, and seniors: long-term results, survival, and adverse events in 1000 consecutive patients. Transplant Proc 1998; 30(4): 1403.
6. Jain A, Reyes J, Kashyap R, et al. What have we learned about primary liver transplantation under tacrolimus immunosuppression? Long-term follow-up of the first 1000 patients. Ann Surg 1999; 230(3): 441.
7. Mazariegos GV, Reyes J, Marino I, Flynn B, Fung JJ, Starzl TE. Risks and benefits of weaning immunosuppression in liver transplant recipients: long-term follow-up. Transplant Proc 1997; 29(1–2): 1174.
8. Stegall MD, Wachs ME, Everson G, et al. Prednisone withdrawal 14 days after liver transplantation with mycophenolate: a prospective trial of cyclosporine and tacrolimus. Transplantation 1997; 64(12): 1755.
9. Gomez R, Moreno E, Colina F, et al. Steroid withdrawal is safe and beneficial in stable cyclosporine-treated liver transplant patients. J Hepatol 1998; 28(1): 150.
10. Everson GT, Trouillot T, Wachs M, et al. Early steroid withdrawal in liver transplantation is safe and beneficial. Liver Transpl Surg 1999; 5(4 Suppl 1): S48.
11. Jain AB, Todo S, Fung JJ, et al. Correlation of rejection episodes with FK 506 dosage, FK 506 level, and steroids following primary orthotopic liver transplant. Transplant Proc 1991; 23(6): 3023.
12. Todo S, Fung JJ, Demetris AJ, Jain A, Venkataramanan R, Starzl TE. Early trials with FK 506 as primary treatment in liver transplantation. Transplant Proc 1990; 22(1): 13.
13. Tisone G, Angelico M, Palmieri G, et al. Immunosuppression without prednisone after liver transplantation is safe and associated with normal early graft function: preliminary results of a randomized study. Transpl Int 1998; 11 (Suppl 1): S267.
14. Padbury RT, Gunson BK, Dousset B, et al. Steroid withdrawal from long-term immunosuppression in liver allograft recipients. Transplantation 1993; 55(4): 789.
15. Punch JD, Shieck VL, Campbell DA, Bromberg JS, Turcotte JG, Merion RM. Corticosteroid withdrawal after liver transplantation. Surgery 1995; 118(4): 783.
16. Ramirez CB, Sebayel MI, Kizilisik T. Early steroid withdrawal after liver transplantation. Transplant Proc 1998; 30(7): 3182.
17. McDiarmid SV, Farmer DA, Goldstein LI, et al. A randomized prospective trial of steroid withdrawal after liver transplantation. Transplantation 1995; 60(12): 1443.
18. DeCarlis L, Belli LS, Rondinara GF, et al. Early steroid withdrawal in liver transplant patients: final report of a prospective randomized trial. Transplant Proc 1997; 29(1–2): 539.
19. Mazariegos GV, Reyes J, Marino IR, et al. Weaning of immunosuppression in liver transplant recipients. Transplantation 1997; 63(2): 243.
20. Starzl TE, Demetris AJ, Murase N, Thomson AW, Trucco M, Ricordi C. Donor cell chimerism permitted by immunosuppressive drugs: a new view of organ transplantation. Trends Pharmacol Sci 1993; 14(5): 217.
21. McMaster P, Gunson B, Min X, Afonso R, Bastos J. Liver transplantation: changing goals in immunosuppression. Transplant Proc 1998; 30(5): 1819.

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IMPACT OF DONOR-SPECIFIC ANTIBODIES ON CHRONIC REJECTION OCCURRENCE AND GRAFT LOSS IN RENAL TRANSPLANTATION: POSTTRANSPLANT ANALYSIS USING FLOW CYTOMETRIC TECHNIQUES[1]

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Background. **Improvements in immunosuppressive therapy have greatly reduced acute rejection (ARj) episodes, ensuring better short-term graft outcome, but have not modified long-term survival in renal transplantation. It is now well accepted that chronic rejection (CRj) can be determined by both immune and/or nonimmune mechanisms. The aim of this study was to evaluate the importance of the posttransplant humoral immune response towards mismatched HLA graft antigens in CRj occurrence and graft outcome.**

Methods. **Serum samples from 120 nonpresensitized renal transplant recipients were prospectively screened for 1 year after surgery by means of flow cytometry cross-match (FCXM) and FlowPRA beads (microbeads coated with purified HLA class I and class II antigens) assays. All transplants were followed-up for 2 years or until graft removal.**

Results. **FCXM monitoring identified donor-specific**

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