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Liver, Kidney, and Thoracic Organ Transplantation Under FK 506

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The new immunosuppressive drug FK 506 was used from the outset with low doses of prednisone to treat 120 recipients of primary liver grafts and 20 more patients undergoing liver retransplantation. The patient survival rate after 2 to 8 months in the primary liver transplantation series is 93.3%, with original graft survival of 87.5%. Of the 20 patients in the hepatic retransplant series, 17 (85%) are living. Almost all of the surviving patients have good liver function. In addition 11 hearts, 2 double lungs, and a heart–lung have been transplanted under FK 506, with survival of all 14 patients. With all of the organ systems so far tested, including the kidney (which has been reported elsewhere), rejection usually has been controlled without additional drugs and with lower average steroid doses than in the past. Nephrotoxicity has been observed, but not to an alarming degree, and there has been a notable absence of hypertension. There is a suggestion that serum cholesterol may be lowered by FK 506, but this is unproved. Although the adverse reactions of FK 506 and the immunosuppressive mechanisms resemble those of cyclosporine, our preliminary observations suggest that FK 506 may have a more advantageous therapeutic index.

THE NEW IMMUNOSUPPRESSIVE drug FK 506 was introduced clinically in February 1989 to replace cyclosporine for liver recipients who had intractable rejection or drug toxicity.[1,2] In March 1989, it was first used as primary therapy from the time of a cadaveric kidney transplantation.[1] Between then and the beginning of February 1990, 140 liver and 36 kidney recipients had grafting procedures at the Presbyterian University and Children's Hospital of Pittsburgh under therapy with FK 506 and prednisone from the time of transplantation.[3,4] In addition 14 patients had transplantation of the heart, lungs, or heart–lung. The use of FK 506 for multiple-organ transplantation, pancreatic islet cell transplantation, and as a device to facilitate the recent development of a separate Veterans Administration program will be reported separately.

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Methods

Liver Transplantation

The 140 patients entered into the study between July 2, 1989 and February 4, 1990 were heterogenous in their indications for transplantation, degree of illness, and age. For analysis they were stratified into the 120 patients receiving their first liver (primary transplantation) and 20 who were receiving their second to sixth liver (retransplantation). Each main group was subdivided into adult (18 years or older) and pediatric (younger than 18 years) categories. The techniques of liver transplantation have been reviewed elsewhere.[5]

Primary Liver Transplantation (120 Cases)

There were 105 adults and 15 infants and children (Table 1) who were given a total of 132 livers. The average adult age of 45 ± 11.5 (SD) years reflected both the aging population in our region and our nondiscriminatory policy toward older candidates; 13 (10.8%) of the 105 adults were 60 years or older. Nonalcoholic and alcoholic cirrhosis accounted for two thirds of the adult indications; 9 of these patients were B-virus (HBV) carriers. The technically 'easier' cholestatic diseases, which include primary biliary cirrhosis and sclerosing cholangitis, were only 27% of the total. There was a generally high degree of illness and urgency as this was defined by the current United Network for Organ Sharing (UNOS) stratification (Table 1): status 1, at home, functioning without nursing care; status 2, at home, not working and requiring professional nursing care; status 3, hospital bound; status 4, ICU bound; UNOstat, ICU bound on life support. Only three of the adult UNOstat patients had fulminant hepatic failure. Many of the patients with chronic liver disease had gravitated to the desperate UNOstat condition over a long period while they waited for a liver to become available.

TABLE 1. *Clinical Features of Primary Liver Recipients*

Feature	Adult (≥18 years)	Pediatric (<18 years)
Number of patients	105	15
Number of transplants	116	16
Age (years)*	45 ± 11.5 (18 to 69)	4.1 ± 4.8 (0.4 to 16)
Sex (M/F)	58/47	9/6
Disease category: No. of cases (%)		
Nonalcoholic cirrhosis†	42 (40)	Biliary atresia 8 (53.3)
Alcoholic cirrhosis	26 (24.7)	Cirrhosis 4 (26.7)
Cholestatic disease	28 (26.6)	Tumor 2 (13.3)
Tumor	3 (2.9)	Fulminant failure 1 (6.7)
Fulminant failure	3 (2.9)	
Miscellaneous	3 (2.9)	
Total	105 (100)	15 (100)
UNOS score: No. of cases (%)		
2	25 (23.8)	5 (33.3)
3	39 (37.2)	5 (33.3)
4	14 (13.3)	3 (20.0)
UNOSTAT	27 (25.7)	2 (13.4)

* Thirteen patients were ≥ 60 years old; 8 were younger than 2 years old.

† Included 9 B-virus (HBV) carriers.

Biliary atresia was the native liver disease in 8 of the 15 pediatric recipients, with cirrhosis a distant second (Table 1). The youngest child was 4.5 months old. The average degree of urgency in these patients was less than in the adults.

Liver Retransplantation (20 Cases)

These 20 patients (15 adult and 5 pediatric) experienced failure of one to five previous grafts that had been in place for 2 days to 80 months under cocktail immunosuppression with cyclosporine and prednisone, with or without azathioprine and/or antilymphoid globulin (ALG) agents (Table 2). The main reasons for graft failure were chronic rejection and hepatic artery thrombosis (Table 2). Several of these patients were sent to Pittsburgh for salvage after having their first transplantation(s) at other centers.

TABLE 2. *Circumstances of Liver Retransplantation (ReTX)*

Circumstance	Adult (≥18 years)	Pediatric (<18 years)
Number of patients	15	5
Number of transplants	18	5
Age (years)	45 ± 4.5 (21 to 64)	5.6 ± 6.5 (1 to 17)
Sex (M/F)	7/8	2/3
Causes of ReTx:		
No. of cases and % ( )		
Rejection	8 (53.3)	5 (100)
HA thrombosis	4 (26.7)	
Primary dysfunction	2 (13.3)	
Recurrent HBV	1 (6.7)	
Number of grafts		
2	10	4
3	4	1
6	1	
Days from		
First graft	728 ± 862 (2 to 2435)	478 ± 278 (84 to 862)
Previous graft	460 ± 594 (2 to 2421)	475 ± 251 (84 to 862)

Treatment of the 20 patients required 23 grafts because additional retransplantation became necessary in three instances.

Thoracic Organ Transplantation

Heart transplantation. Between October 8, 1989 and February 14, 1990, heart transplantations were performed in eight adults (48 to 56 years) and three children (who were 11 months, 2.5 years, and 11 years old). The indications were idiopathic cardiomyopathy (n = 5), ischemic cardiomyopathy (n = 5), and congenital heart disease (n = 1). Four of the 11 patients needed preoperative mechanical assist device support and three more required inotropic drugs. The donor for a 52-year-old woman was her 48-year-old sister (one haplotype HLA match) who died of a stroke; special permission was obtained from UNOS to permit allocation outside of the normal priority list.

Lung or heart–lung transplantation. Two men with cystic fibrosis who were 34 and 27 years old had double-lung transplantation on October 13 and November 28, 1989, respectively. A 27-year-old woman with Eisenmenger complex had a heart–lung transplantation on October 20, 1989. She had minimal residual disability from a cerebrovascular accident several years earlier, which presumably was related to polycythemia.

Kidney Transplantation

Thirty-six kidney transplantations were performed between March 27, 1989 and January 4, 1990 and have been reported elsewhere.[4] More than two thirds of these patients were not conventional kidney transplant recipients because of previous or concurrent transplantation of

the liver (n = 10) or heart (n = 1), positive cytotoxic cross-matches (n = 9), and other complicating factors that normally would have precluded renal transplant candidacy. Two (5.6%) of the patients died. Twenty-seven (75%) have good graft function (mean serum creatinine, 1.7 mg%) after 3 to 13 months follow-up (156 ± 52 [SD] days). In the present report, these cases were used for comparison of FK 506 doses, FK 506 plasma levels, and cholesterol concentrations with those of primary liver and thoracic organ recipients.

Immunosuppression

FK 506. Initial treatment is the same with all organ recipients and has been standardized as follows. Intravenous doses of 0.075 mg/kg infused during 4 hours are started in the operating room and repeated every 12 hours until oral intake is begun. The conversion from intravenous to oral doses of 0.15 mg/kg every 12 hours usually is overlapped for 1 day. Dose revisions are made on clinical grounds and guided by trough plasma levels of FK 506, which are measured with an enzyme immunoassay technique.[6] Optimal 12-hour trough levels are thought to be in the 0.5 to 1.5 ng/mL range, but concentrations less than this may be adequate, and higher concentrations may be well tolerated.[4] Most upward dose adjustments are responses to the break-through of rejection, combined with low plasma levels of FK 506. Downward dose adjustments usually are dictated by adverse drug reactions of which neurotoxicity is the most common and useful for clinical management.[4] Major nephrotoxicity implies serious overdosage but is quickly reversible.

Later, variations from this generic approach were dictated by the nature of the graft, the quality of graft function, and kidney function in nonrenal and renal recipients. Because the principal metabolism of FK 506 is by the liver, the doses were reduced after hepatic transplantation if the initial liver graft function was substandard. Under these circumstances, downward adjustments of FK 506 dosage were correlated with liver dysfunction, declining renal function, neurotoxic side effects, and the demonstration of supratherapeutic plasma levels of FK 506. These were documented as high as 25 ng/mL. The supreme value of monitoring plasma levels of FK 506 was in the liver recipients. In liver recipients upward adjustments of FK 506 dosage from the generic regimen were rarely made perioperatively and then only if there was unequivocal evidence of rejection. Protocol liver biopsies were obtained at 2 weeks and 2 months to help guide management.

An effort was made in recipients of thoracic organs to keep the FK 506 doses as high as possible. Perioperatively renal dysfunction necessitated temporary reduction of the intravenous doses in one half of the cases. This was most common when circulatory or pulmonary assist procedures had been in effect before operation. Later, changes in immunosuppression were based on the histologic findings in serial endomyocardial or lung biopsies and the results of bronchopulmonary lavage specimens. The initial response to a rejection was to increase the FK 506 dose if renal function was adequate. This approach permitted the double-lung recipients and the heart–lung recipient to be managed without maintenance steroids or any other adjuvant agent.

Kidney recipients were managed in the same general way as heart patients. Elevations of creatinine were evaluated with standard radiologic studies and kidney biopsies were obtained as needed to establish the differential diagnosis of rejection *versus* nephrotoxicity or ischemic injury. In such cases FK 506 doses were increased if there was evidence of rejection with little or no toxicity, or reduced if the biopsy did not show rejection.

Other drugs. All of the patients reported here, except for the lung and heart–lung recipients, also were given prednisone. In the first part of the experience, 1 g intravenous methylprednisolone was given to adults during operation, and a 5-day burst of methylprednisolone was begun at 200 mg on the first day and reduced daily in 40-mg steps. The doses were scaled down for infants and children. Beginning in late 1989, the intraoperative bolus and subsequent steroid cycle were omitted. Instead a daily dose was started of 20 methylprednisolone. Prednisone doses were reduced to 0 to 10 mg/day during the second to sixth post-transplantation weeks if there was no evidence of rejection.

Rejection episodes that were unresponsive to increasing the maintenance doses of FK 506 were treated with a single 1-g bolus of methylprednisolone or hydrocortisone in adults or with lesser quantities in children. If rejection persisted additional steroids were given, a 3- to 5-day course of 5 or 10 mg/day OKT3 was considered, and in a few cases azathioprine was added.

Tissue Matching

As would be expected with random matching, poor HLA compatibility was present with all of the extrarenal transplantations, except for the sister-to-sister heart transplantation. Efforts at matching in the renal cases were completely unsuccessful, despite considerable effort.[4] Twelve (10%) of the 120 primary liver transplantations were performed, despite completely killing positive cytotoxic cross-matches. All but one of these recipients had a panel reactive antibody index (PRA) ≥ 40%. None of the thoracic transplant recipients had circulating cytotoxic antibodies.

Data Analysis

Patient and graft survival were recorded for each organ and actuarial survival was projected with Kaplan–Meier analyses. Data compilations were expressed as mean ± standard deviations (SD). Data from our recently reported FK 506-treated kidney recipients were used for comparisons of the FK 506 doses, blood levels, and cholesterol levels with those in recipients of other organs. Comparisons were made with a one-tailed student's t test and X^2 test with continuity correction and were considered significant at $p < 0.05$.

For comparison of the present results with our previous liver transplantation experience, 400 consecutive primary transplant cases were studied, going back from November 8, 1988 to October 25, 1987. Then, as during the FK 506 study, all livers were preserved with University of Wisconsin (UW) solution. The historical controls had about the same adult and pediatric case representation as during the FK 506 study. Historical controls from November 1988 onward were not useful because so many of the later patients eventually were switched from cyclosporine to FK 506.[2] In addition to patient and graft survival in all 400 cases, several other biochemical and treatment parameters were examined in smaller numbers that were determined by data availability.

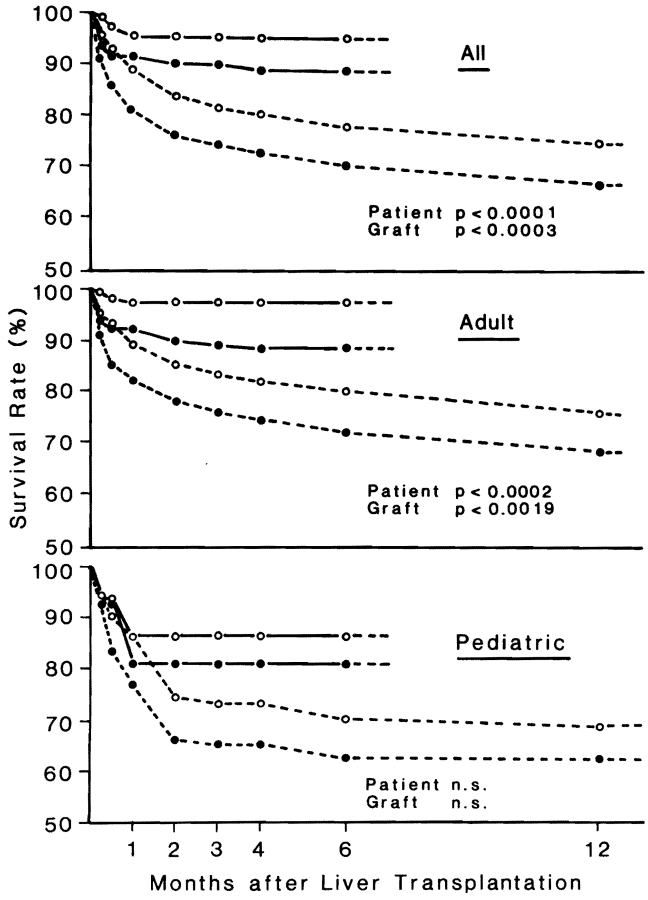

FIG. 1. Patient (○) and graft (●) survival rates in 120 cases under FK 506 therapy (——) and in 400 historical controls (– – –) after primary liver transplantation in all patients (upper), adults (middle), and pediatric recipients (lower).

TABLE 3. *Causes of Retransplantation (RTx) Within 60 Days After Primary Liver Transplantation Under FK 506* Versus *Historical Controls*

	FK 506*	Cyclosporine	p
N (Starting)	120	400	
N (RTx)	7 (5.8%)	60 (15%)	<0.05
Reasons for RTx			
Rejection	1 (0.8%)	15 (3.8%)	—
Artery thrombosis	1 (0.8%)	9 (2.2%)	—
Primary dysfunction	5 (4.2%)	25 (6.2%)	—
Viral hepatitis	0 0	6 (1.5%)	—
Other	0 0	5 (1.3%)	—

Outcome of second transplantation: Five of the seven FK 506 patients are alive 3 to 5.5 months later. One of the patients who died received a total of three livers. The 90-day survival rate after retransplantation in the cyclosporine group was 34 of 60 patients.

* The patient and graft survival curves up to and beyond 60 days after primary transplantation are shown in Figure 1. Two more patients required retransplantation after 60 days in the FK 506 series, and both patients survived after receipt of their second graft and in one case a subsequent third graft.

Results

Primary Liver Transplantation

Patient and graft survival. One hundred twelve (93.3%) of the 120 patients are alive after 2 to 7.5 months. The patient ($p < 0.0001$) and graft ($p < 0.0003$) survival was superior to that in the historical controls (Fig. 1, top). This advantage was seen both in the adult (Fig. 1, middle) and pediatric cases (Fig. 1, bottom), although the numbers did not reach significance in the infants and children.

Rate of retransplantation. The diminished need for retransplantation at all times under FK 506 therapy is evident in Figure 1 and is documented in Table 3 for the first 60 days. Seven retransplantations (5.8%) were performed, an incidence that was almost one third of the 15% in the historical control group ($p < 0.05$). All of the usual causes of retransplantation shared in the improvement (Table 3). Also noteworthy was the absence of graft losses from viral infection.

The only FK 506-treated graft that was lost to rejection was in a highly sensitized patient who had a PRA of 100%, and completely killing positive antidonor cytotoxic cross-matches. This liver was not hyperacutely rejected (Fig. 2), but intense lymphocytic infiltration was found on biopsies 7 and 29 days after operation (Fig. 2). A second liver was transplanted after 40 days, also against positive cross-matches. This graft eventually provided normal function under intensified immunosuppression (Fig. 2)

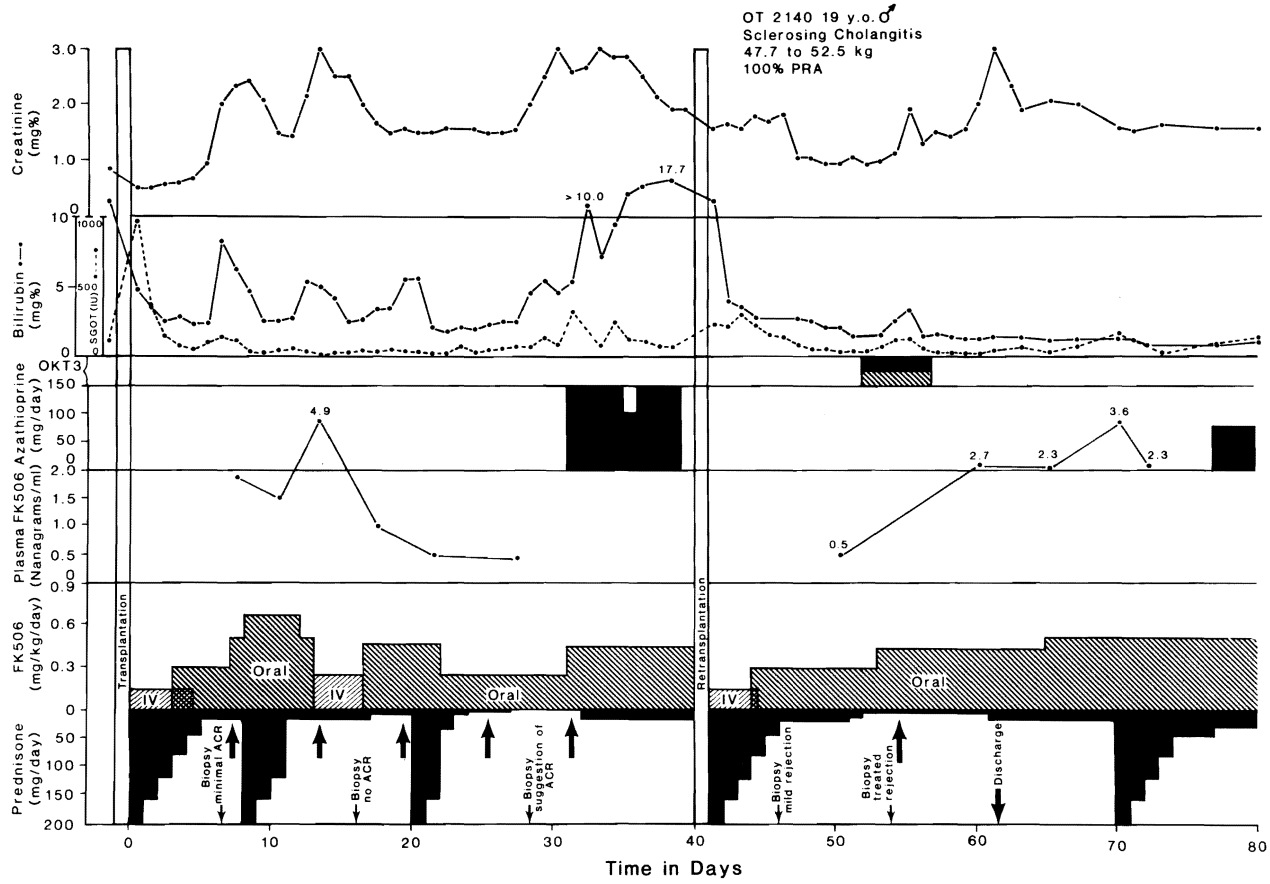

FIG. 2. Difficult course of a highly sensitized patient (PRA, 100%) who received a liver across a 100% kill cytotoxic cross-match. The therapeutic adjustments were inadequate during the rejection of the first graft. After retransplantation, increased steroids, a course of OKT3, and azathioprine were added. Eventually satisfactory function could be sustained with FK 506 and prednisone doses of 30 mg/day. The patient now is 7 months after operation and has had prednisone reduced to 5 mg/day. Note the creatinine rise, which occurred as FK 506 was increased to supernormal levels. Large upward arrows = steroid boluses. ACR, acute cellular rejection.

but at the cost of nephrotoxicity from high-dose FK 506 therapy.

It may be significant that three of the retransplantations within 60 days in the FK 506 series were in patients with strongly positive cytotoxic cross-matches. These early graft losses were from the total of 12 cases in which livers were confronted with such cross-matches. The 60-day graft loss of 25% in the cross-match–positive cases *versus* 3.7% (4 of 108) in the cross-match–negative cases was significant ($p < 0.05$).

Causes of death. Eight (6.7%) of the 120 primary liver recipients died within 60 days for the reasons summarized in Table 4. There have been no deaths after 60 days. The 60-day mortality rate was significantly less than in the historical controls ($p < 0.05$). Two of the deaths were after emergency retransplantation. Sepsis caused 4 of the 8 deaths. However lethal sepsis occurred at a much reduced rate compared to the historical controls ($p < 0.05$).

The fatal intraoperative complication (Table 4) was caused by an unrecognized laceration of the subclavian artery in a 4-year-old child during anesthesia induction and placement of a central line. The patient was given only a partial first dose of FK 506 before developing an

TABLE 4. *Causes of 60-day Mortality* After Primary Liver Transplantation (FK 506* Versus *Historical Controls)*

	FK 506	Cyclosporine	p
N (starting)	120	400	
N (deaths)	8 (6.7%)	66 (16.5%)	<0.05
Causes of death			
Sepsis	4 (3.3%)	40 (10%)	<0.05
Intraoperative	1 (0.8%)	7 (1.8%)	—
Liver failure	0 (0%)	6 (1.5%)	—
Heart failure	1 (0.8%)	4 (1%)	—
Stroke	1 (0.8%)	1 (0.2%)	—
Others	1† (0.8%)	8 (2%)	—

* There have been no deaths after 60 days in the FK 506 group. The continuing mortality in the historical controls is evident in Figure 1.

† Fulminant failure: failed to wake up.

acute hemothorax. Another child with fulminant hepatic failure did not wake up after the transplantation. Because of our policy of agressive retransplantation, the incidence of death from hepatic failure was low in both the FK 506 and historical cases.

Liver function. One hundred twelve of the starting 120 primary liver recipients are alive, 7 after retransplantation (5 before 60 days and 2 after 60 days; Table 3). The function of the 105 grafts that have been in place continuously for 2 to 8.5 months is shown in Table 5. Only one patient with a recent biliary reconstruction has a bilirubin level more than 2 mg%. The serum glutamic oxaloacetic transaminase and alkaline phosphatase values are also normal or only slightly elevated in almost all cases.

The seven other patients who are alive after retransplantation also have generally good liver function, with only one exception.

Steroids, rejection, and adjuvant immunosuppression. Most FK 506 patients can be weaned from the prednisone rapidly during the first month (Figs. 3 and 4), leaving the FK 506 survivors on an average prednisone dose of only 6.6 mg/day at the end of this time compared to 16.7 mg in the historical controls ($p < 0.05$). Further steroid reductions were routinely made so that by 2 and 3 months the daily average doses were much less than 5 mg, significantly less than in the historical controls (Fig. 5). The appreciation of the inter-relationship between rejection and all modalities of immunosuppression during the first 30 days can be gained from Table 6. The absolute rate of clinical rejection was less ($p < 0.01$) in the FK 506-treated patients than in the historical controls, but only slightly less (NS) by histopathologic criteria. At the same time, the FK 506 patients were given only 43% of the steroid boluses used in the cyclosporine controls, 7% of the 5-day steroid recycles, 4% of the azathioprine, and 23% of the OKT3 courses.

TABLE 5. *Present Metabolic Status of 105 Liver Recipients (87.5% of Original 120) Whose Primary Transplantations Were August 17, 1989 to February 4, 1990. Data from Seven Other Surviving Primary Recipients who Required Retransplantation Are Not Included (Mean ± SD).*

	N	
105 Days survival		128 ± 46
*105 Bilirubin (mg%)		0.8 ± 1.3
105 SGOT (IU/L)		50.3 ± 57.1
†92 Alkaline phosphatase (IU/L)		153 ± 136
†92 Serum cholesterol (mg%)		160 ± 53
95 Uric acid (mg%)		6.9 ± 4.2
105 BUN (mg%)		24.5 ± 12.3
105 Creatinine (mg%)		1.4 ± 0.7
61 Magnesium (mg%)		1.3 ± 0.3

Normal values of: cholesterol: 130 to 240 mg% (age dependent); uric acid: <8.5 mg% males, <7 mg% females; magnesium: 1.8 to 2.4 mg%.

* Only one patient with recently repaired duct obstruction has a bilirubin ≥ 2 mg%.

† Adults only.

SGOT, serum glutamic oxaloacetic transaminase.

BUN, blood urea nitrogen.

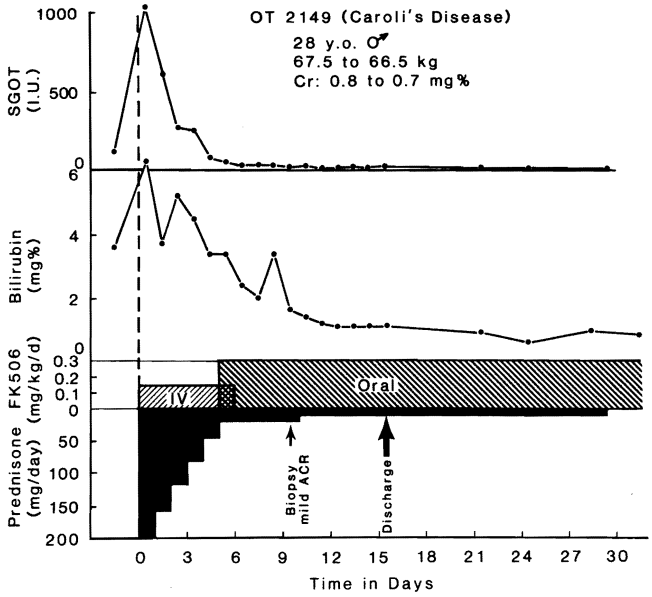

FIG. 3. Benign convalescence after liver transplantation in a 28-year-old man. The biopsy at 12 days showed mild acute cellular rejection (ACR) for which no treatment was given. He is well after 7 months. Cr = serum creatinine. SGOT, serum glutamic oxaloacetic transaminase.

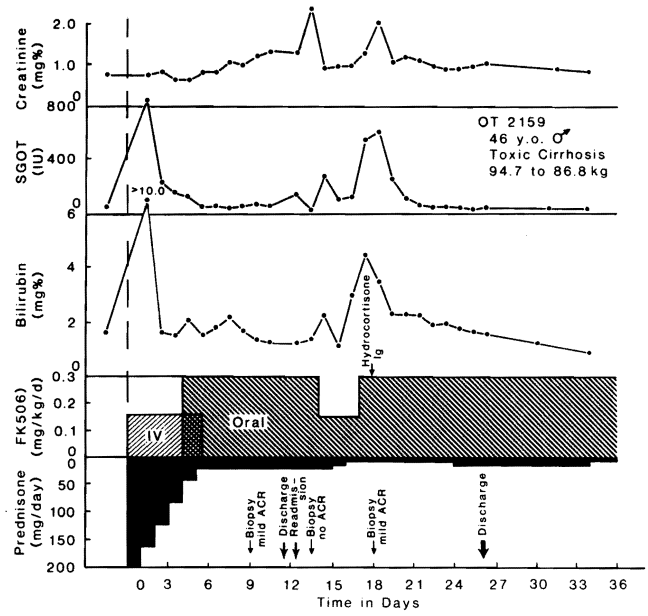

FIG. 4. Treatment of a liver rejection with a single 1-g bolus of hydrocortisone in a patient whose FK 506 was reduced temporarily because of a minor elevation of serum creatinine. After restoration of the FK 506 dose and a small increase in baseline steroids, normal function was restored. The patient is well after 7 months. ACR, acute cellular rejection. SGOT, serum glutamic oxaloacetic transaminase.

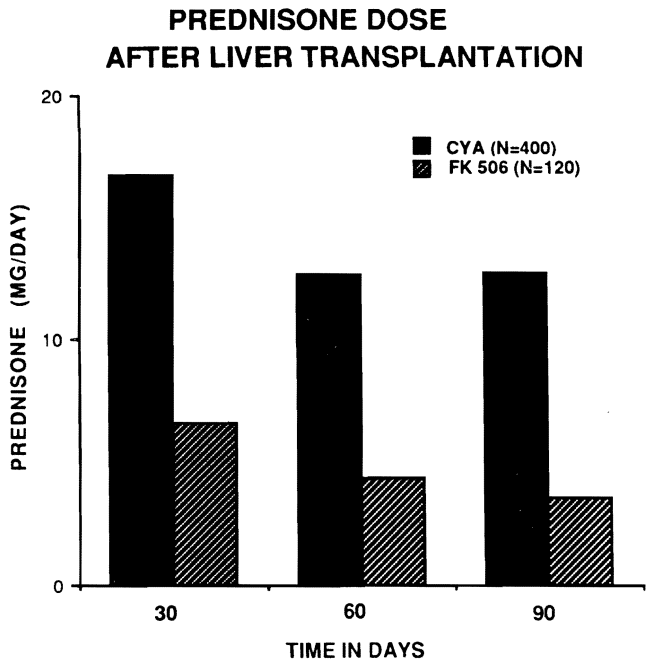

FIG. 5. Daily prednisone after 1, 2, and 3 months in the primary liver recipients treated with FK 506 *versus* the historical controls.
CYA, cyclosporine.

The simplified management made possible by FK 506 in most patients is seen in Figures 3 and 4. Rapid recovery, early discharge from the hospital, and steroid discontinuance were possible in more than one third of the cases (Fig. 3). The patient whose course is summarized in Figure 4 had a significant rejection that was easily reversed with a single steroid bolus and an increase of the FK 506 dose.

Other metabolic tests in FK 506 patients. Present average renal function in the 105 liver recipients still bearing their original hepatic grafts is nearly normal (Table 5), as judged by serum creatinine and blood urea nitrogen. Current serum cholesterol, serum uric acid, and serum magnesium in the same patients are summarized in Table 5.

Infectious complications. The reduced incidence of fatal infections was documented in Table 4. However the patterns of nonlethal infectious disease were similar to other immunosuppressive regimens and included wound infections requiring treatment with antibiotics or surgical drainage, urinary tract infections, pneumonias, and fungal infections. The most common virus infection was cytomegalovirus. There were no protozoan infections.

Lymphoma. Polyclonal lymphoproliferative disease developed in two patients who had Epstein–Barr virus infections. A 19-year-old man had an infectious mononucleosis syndrome with enlargement of tonsils. After tracheostomy, tonsillectomy, reduction of the FK 506 dose, and treatment with acyclovir, he recovered and has no residual effects 5 months later. A 65-year-old woman with a PRA of 100% and a positive cytotoxic cross-match rejected both her primary and a second liver, despite additional therapy with high doses of prednisone, multiple courses of OKT3, and azathioprine. She died of sepsis 9 days after a third transplantation. The second graft contained a polyclonal lymphoma with no other foci.

Adverse reactions. Two of the 120 primary liver recipients developed transient expressive aphasia 6 and 11 days after transplantation. These cases have been reported in detail.[7] Other major neurologic complications, such as convulsions and coma, were not seen. One patient who died of cerebral hemorrhage 4 days after operation had thrombocytopenia (platelet count 10,000). Minor neurotoxic symptoms such as tremors, paresthesias, increased sensitivity to light, insomnia, and mood changes were common and were helpful in guiding dose adjustments.[4]

Ten of the one-hundred twenty primary liver recipients developed new-onset diabetes requiring insulin. If only the 112 survivors are considered, the incidence was 8.9%. The incidence of new diabetes in kidney recipients treated with cyclosporine regimens has been reported to be 10% to 20%.[8,9]

There were no examples of hirsutism, gingival hyperplasia, or gynecomastia. A few patients noted thinning of the hair.

Liver Retransplantation

These 20 patients (15 adults and 5 children) had been under treatment that included cyclosporine and prednisone. Seventeen (85%) are alive with follow-ups of 2.3 to 9 months (Fig. 6). Two of the three patients who eventually

TABLE 6. *Other Immunosuppressive Agents and Rejection in Patients Within 30 Days After Liver Transplantation*

		FK	CyA	p<
N		120	303	
Steroid bolus	No	77 (64.2)	51 (18.8)	
	Yes	43 (35.8)	252 (81.2)	0.001
Steroid recycle	No	117 (97.5)	201 (66.3)	
	Yes	3 (2.5)	102 (33.7)	0.001
Immuran	No	117 (97.5)	113 (37.3)	
	Yes	3 (2.5)	190 (62.7)	0.001
OKT3	No	107 (89.2)	159 (52.5)	
	Yes	13 (10.8)	144 (47.5)	0.001
Rejection				
Clinical	No	73 (60.8)	138 (45.5)	
	Yes	47 (39.2)	165 (54.5)	0.01
Histopathologic*	No	51/106 (48.1)	104/248 (41.9)	
	Yes	55/106 (51.9)	144/248 (58.1)	NS

* The pathology data were not obtained under comparable circumstances. All of the adult FK 506 patients had protocol biopsies at 2 weeks and 2 months, whereas the livers in the historical control cases were sampled irregularly. A valid comparison of histopathology will require a randomized trial and protocol biopsies in both limbs.
NS, not significant.

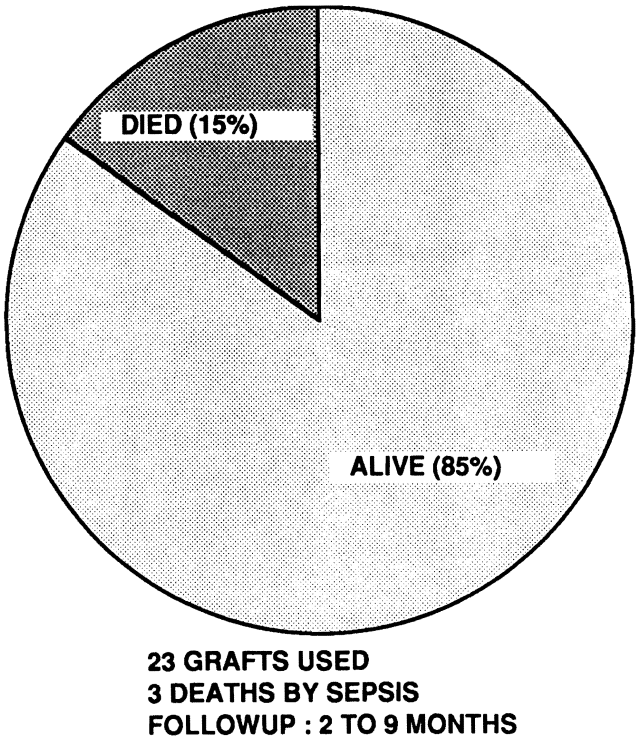

FIG. 6. Survival of patients whose previous grafts were lost under cyclosporine regimens and who underwent retransplantation with FK 506 treatment.

died of sepsis had a further attempt at retransplantation, in one patient twice, accounting for a total use of 23 grafts to treat these 20 recipients. The third death was in a child with perfect graft function who had a respiratory arrest on the ward, probably because the transplanted liver was too large for the body size.

The 17 patients who survived had more infections, a higher incidence of renal failure, and longer hospitalizations than did the patients in the primary series. However liver graft function at 30 days and subsequently was equivalent to that in the primary transplantation series. One of the survivors was a child who underwent pancreaticoduodenectomy because of pre-existing pancreatitis that became hemorrhagic after operation. Pre-existing hirsutism and gingival hyperplasia in patients who had been on chronic cyclosporine therapy reversed in 1 to 2 months.

Transplantation of Thoracic Organs

A detailed report of these cases is scheduled for presentation to the American Society of Transplant Surgeons in June 1990 and the International Transplantation Society in August 1990. The two double-lung and the heart–lung recipients are well after 130, 165, and 173 days, respectively, never having received maintenance steroid therapy. One of the double-lung recipients has been suspected to have bronchiolitis obliterans because of biopsy findings. Because he has no complaints or deterioration of pulmonary function, other maintenance immunosuppression has not been added to FK 506.

The 11 heart recipients also are well after a mean survival time of 104 ± 54 (SD) days (51 to 180 days). One adult was treated with OKT3 and started on azathioprine as a third maintenance drug after 3 months because of histopathologic evidence of rejection. An 11-year-old child was switched by his pediatric cardiologist from FK 506 to azathioprine after 3 months, probably with insufficient reason. Other potentially important information is summarized in Table 7. The patients have been unusually free of the hypertension that was common in the historical controls. Serum creatinine concentrations were approximately the same in both groups. The serum cholesterol levels are less than in the historical controls but not significantly so.

Kidney Transplantation

Current data on the 27 kidney recipients with functioning grafts are summarized in Table 8. The low steroid

TABLE 7. *Parameters in Surviving Heart Transplantation Recipients*

Parameter	FK 506			Cyclosporine		
Days	0	30	51–90	0	30	51–90
n	11	11	11	40	36	35
Prednisone (mg/day)	—	13.6 ± 7.2	10.7 ± 8.5	—	17.8 ± 4.7	18.0 ± 4.5
Antihypertensive drugs						
One	—	0/11	1/11	—	11/36	19/35
Two	—	0/11	0/11	—	2/36	1/35
Total	—	0/11	1/11 (9%)	—	13/36 (36%)	20/35 (57%)
Serum cholesterol (mg%)		210 ± 52	220 ± 52		216 ± 41	245 ± 55
Serum creatinine (mg%)		1.1 ± 0.6	1.4 ± 0.8		1.0 ± 0.4	1.2 ± 0.5

TABLE 8. *Present Metabolic Status of 27 Kidney Recipients Whose Primary Transplantations Were March 27, 1989 to January 4, 1990**

	Renal Recipients	Normal Values
Mean number of days after transplant	156 ± 52	
Creatinine (mg%)	1.75 ± 0.75	0.5–1.4
Blood urea nitrogen (mg%)	29.3 ± 13.8	5–20
Uric acid (mg%)	7.0 ± 2.3	2.5 ± 8.5
Magnesium (mg%)	1.5 ± 0.3	1.3 ± 2.1
Serum cholesterol (mg%)	176 ± 44	160–250
Patients on antihypertensives	11/28	
Daily prednisone (mg)†	4.3 ± 6.2	

* Details of all patients in this complex series are reported elsewhere.[4]
† Twenty of the 28 patients are receiving no steroids; four are on 5 mg/day.

doses, good average renal function, relative freedom from hypertension, and low serum cholesterol levels are good prognostic signs that have prompted the institution of a randomized trial in more conventional renal candidates.

Figure 7 shows a comparison of the serum cholesterol in these renal recipients with those in the liver and heart recipients. The lowest values were in the liver recipients, and the highest were in the heart recipients. The two double-lung and heart–lung recipients had very low cholesterol levels.

A similar interorgan comparison is shown in Figure 8 for FK 506 doses and plasma blood levels. Kidney recipients appeared to require higher doses to maintain plasma levels equivalent to those of the liver patients. The thoracic organ recipients were more heavily treated as a policy and had higher plasma levels.

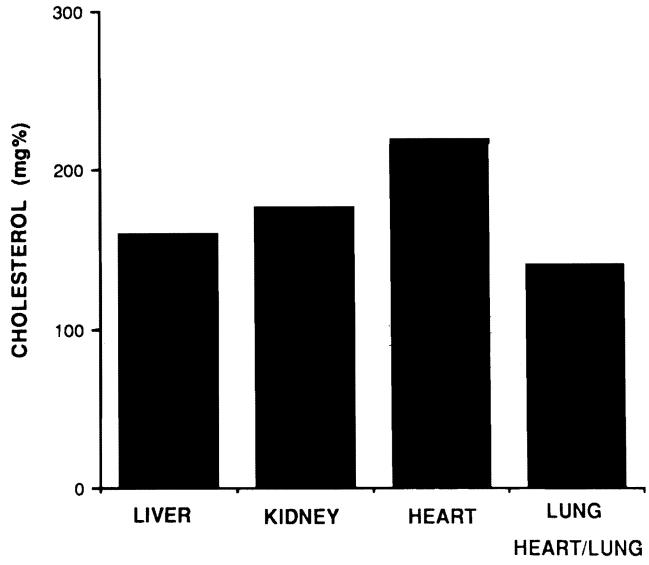

FIG. 7. Serum cholesterol comparison in recipients of different organ recipients.

Discussion

The accomplishments in transplantation during the last 30 years, and exponentially in the last decade, have exceeded expectations. Small steps, widely spaced in time, became large leaps with the advent of cyclosporine. Yet the nephrotoxicity and other side effects of this drug limited its value and stimulated a search for better agents. FK 506 was a product of this search. It was discovered in Japan by Kino et al.[10] in 1984 during systematic screening for drugs with antimicrobial, antineoplastic, or immunosuppressive qualities.

FK 506 is a macrolide antibiotic (such as erythromcyin) that is recovered from the fermentation broth of the soil fungus *Streptomyces tsukubaensis*. FK 506 has no structural similarity to cyclosporine, but it has in common the ability to inhibit the synthesis and expression of interleukin-2 and other cytokines, including gamma interferon.[10–12] Like cyclosporine, FK 506 suppresses T-lymphocyte activation, adoptive immunity, and alloreactivity (rejection) *in vitro* and in homotransplantation models. The drug can mitigate or prevent the rejection of heart, liver, kidney, pancreas, lung, intestine, and skin grafts in mice, rats, dogs, monkeys, baboons, and humans. The international exchange of this burgeoning information was facilitated by satellite symposia of the European Society of Organ Transplantation in 1987 and 1989.[13,14] Clinical trials have been in progress in Pittsburgh since February 1989. The ultimate role of FK 506 in transplantation practice has not been fully delineated, but there are indications from our experience that it may be a large one.

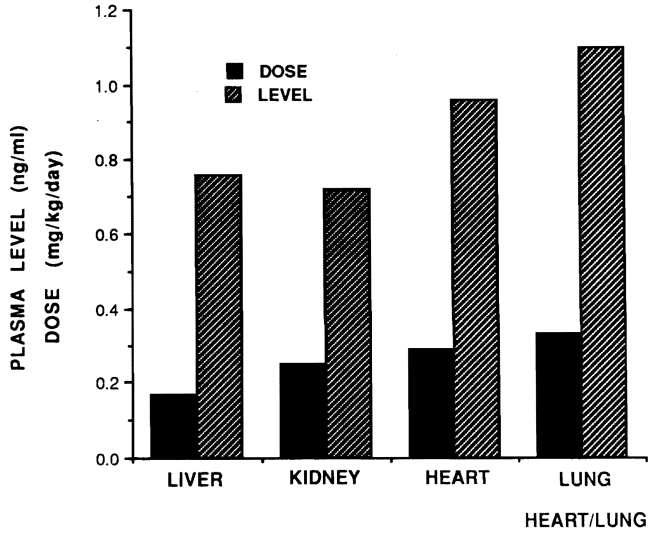

FIG. 8. Dose and plasma level of FK 506 in different organs.

FK 506 was used first clinically to treat liver recipients whose convalescence was unsatisfactory under cyclosporine-containing regimens.[1,2] However all of the transplantations in the present report were with FK 506 therapy from the outset.[3] Because of the variety of the organ grafts, specific questions could be addressed about toxicity, dose requirements, and metabolic changes that might have been less accessible with only one kind of organ graft.

With both primary hepatic transplantation and retransplantation, there were high rates of graft and recipient survival. The patients treated were thought to be of the same level of difficulty if not greater than in our past experience. Extremely ill, old, or very young patients were represented in large numbers as well as those with hepatitis B virus infections, renal failure, previous portal-systemic shunts, and multiple upper-abdominal operations.

The improvement in results with FK 506 compared to historical liver transplantation controls began with a greatly reduced need for retransplantation. A distressing statistic in most liver transplantation centers has been a 10% to 20% incidence of primary graft nonfunction (usually dysfunction), which has been explained by undetected pre-existing donor disease, poorly performed procurement, inadequate preservation, or a flawed recipient operation.[15] Despite the validity of these various explanations in some cases, there has been a surprising lack of correlation between recipient outcome; the use of so-called good, mediocre, and poor donors; or the surgeon's perception of his or her technical performance.[15] The low incidence of primary graft dysfunction and a reduced rate of hepatic artery thrombosis in the FK 506 patients may be indirect evidence that many early postoperative graft dysfunctions or thrombotic events are caused by immunologic factors that have not been recognized. In a few cases, there has been strong evidence that this can occur, despite a negative cytotoxic cross-match.[16] In the present report, the cytotoxic cross-match was identified as a special but surmountable problem.

In addition to high survival rates after both primary liver grafting and retransplantation, the quality of liver function was generally good, reflecting excellent control of rejection. This was achieved with low steroid doses and with minimum need for the azathioprine or the ALG preparations that have been used in cyclosporine 'cocktail' regimens. There was a low incidence of nephrotoxicity in the primary recipients not previously exposed to cyclosporine and a somewhat greater incidence in the retransplantation series. The arterial hypertension that has been associated with cyclosporine therapy[17] was very uncommon. Hirsutism and gingival hyperplasia were never seen in fresh cases and disappeared in retransplant recipients when they were switched from cyclosporine to FK 506.

The impression should not be left that FK 506 is free of nephrotoxicity. Nephrotoxicity is merely less than that of cyclosporine, and seemingly more reversible. Nephrotoxicity was uncommon in the liver recipients and was not used as a barometer to establish a dose ceiling. However, in our double-lung and heart–lung recipients in whom steroids and other immunosuppressive adjuncts were avoided, FK 506 was given to the limit imposed by elevations in the serum creatinine to more than 2 mg%.

These results have given a very favorable impression of FK 506. Whether comparisons with historical controls are valid or fair remains to be determined in future randomized trials because excessive immunosuppression, particularly with prednisone, may have been given in the recent control cases. Ten years ago, when cyclosporine was first used with steroids for liver transplantation, prednisone was used sparingly and discontinued early when possible. The results were comparable to those obtained at other centers in later years.[18] By the late 1980s, a worldwide drift had occurred (including at our center) toward complicated multiple-drug management for all kinds of organ transplantation in an effort to minimize cyclosporine nephrotoxicity. Kahan[17] has pointed out that there have never been well-designed randomized trials to test the legitimacy of this now widely accepted practice. Instead of reflecting better immunosuppression when cyclosporine was used with multiple other drugs, much of the improvement in global results after liver transplantation could be due instead to better preservation methods, acquisition of surgeon experience, improvements in infectious disease management, and more discriminating case selection by teams striving to establish credibility. This last factor may be the most important of all in a competitive market place in which survival claims can make or break a program. How profoundly the seriousness of pre-existing disease and other risk factors, including age, can influence the outcome has been demonstrated precisely in patients with primary biliary cirrhosis.[19]

Whatever the merit of these reservations, the advantage of a simple FK 506-steroid management regimen appeared to be the same with other organs than the liver. For example, in the heart recipients, of whom all 11 are alive, neither permanent renal dysfunction nor serious hypertension have been observed during follow-ups of 2 to 6 months. Because cholesterol is thought to be upregulated by cyclosporine,[20] particular attention was paid to serum cholesterol levels in the FK 506-treated patients. Using FK 506, cholesterol levels have been reported to be low to normal in kidney recipients,[4] and these were even lower in the liver, the lung, or heart–lung recipients in the present report (Fig. 7). However cholesterol lowering was not evident in the 11 heart recipients of the present report, possibly because they were being given more steroids than the other organ recipients. Alternatively hypercholesterolemic patients may have been over represented in the heart group of whom five of the 11 had

coronary artery disease. If the cholesterol-lowering qualities of FK 506 can be validated with more experience, cyclosporine-related hypertension and hyperlipidemia, two of the most serious present-day risk factors for coronary artery disease in heart, kidney, or liver recipients may be ameliorated.

FK 506 has additional metabolic effects. In addition to its immunologic and other qualities, which have been discussed, it is diabetogenic to the same extent as cyclosporine, augments hepatic regeneration, and can be neurotoxic. It has become increasingly apparent that many, if not all, of the organ systems and physiologic end points affected by FK 506 are the same as for cyclosporine, although to differing degrees and sometimes in opposite directions.[4,21] This was puzzling at first because FK 506 and cyclosporine have no structural similarities and have different cytosolic binding sites. However now it is suspected that the two drugs may act on an enzyme called *cis-trans* peptidyl-prolyl isomerase (PPIase), which is a principal constituent of the binding sites.[22,23] PPIase, which was discovered in the pig kidney in 1984,[24] is widely distributed in tissues from the lowest to the most developed species, including humans. Although PPIase was known to facilitate the biologically important process of catalysis of oligopeptide bonds and to facilitate protein folding,[24] the physiologic significance of PPIase did not begin to come to light until its possible role in immune modulation was realized.[22,23] We have speculated that the positive as well as adverse immunologic and other wide-ranging effects of FK 506 and cyclosporine result from alteration (probably inhibition) of the PPIase system in many tissues in ways that are not yet understood.[21]

The influence of the route of administration on these diverse effects also is being clarified by observation of patients. Because orally administered FK 506 and cyclosporine are presented in high concentration to the liver, and largely metabolized there, it has been suggested that they might be more effective in protecting liver grafts from rejection than in protecting other organs, such as the heart and kidney, which are beyond this first pass 'filter.'[25] No conclusions are possible about this possibility. The liver recipients have been maintained on smaller average doses of FK 506 and with lower plasma levels than recipients of kidneys or thoracic organs. The average doses have drifted down in liver recipients largely in response to minor complaints of toxicity and because higher doses did not seem necessary for control of hepatic rejection. This may have reflected physician policy rather than any fundamental biologic difference. Alternatively the difference in FK 506 dosaging with the liver *versus* the nonhepatic transplantations might be because of subtle dysfunction of transplanted livers, which would not be a comparable factor of FK 506 drug disposition in heart or kidney recipients who usually have normal native livers.

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Discussion

Dr. Felix T. Rapaport (Long Island, New York): I want to congratulate Tom Starzl for his outstanding presentation as well as for receiving the Association's highest award for scientific achievement. When the early studies of FK 506 were first presented in 1987, it was quite clear that Tom and his group were the only ones who believed that this agent was any good or had any potential whatsoever. We see today the fruition of Tom Starzl's efforts, exhibiting that tenacious faith and boundless energy of his in developing what is clearly a new milestone in immunosuppression.

It is an irony of nature that the one species that seems to be least susceptible to the toxic effects of FK 506 is its ultimate beneficiary, the human subject.

The importance of FK actually may reach far beyond transplantation, as Tom Starzl just noted, using autoimmune disease as one example.

It is quite possible that FK may be a model probe for further study of the molecular aspects of immune responsiveness and cell function.

Along these lines, while FK seems to exert the same immunosuppressive effects as cyclosporine, I was struck, in seeing the manuscript, that Tom was kind enough to show one that there were key differences between these two agents. They have different binding sites on the lymphocyte surface; FK is a hair thinner, using cyclosporine causes hirsutism; FK has no effect on the blood pressure, and may actually even decrease serum cholesterol levels, while cyclosporine produces hypertension and hyperlipidemia. Finally, FK seems to facilitate transplantation in the face of positive cross matches for anti-HLA antibodies, albeit with a 25% failure risk.

One wonders whether this agent, therefore, could not also be of relevance to facilitate ABO-incompatible transplantation, as a potential model for xenotransplantation.

The results that Dr. Starzl lists in his manuscript suggest that it may be possible that even the most violent humoral-mediated rejection might require the participation of cell populations that can be rendered incompetent by an agent such as FK.

I would be very interested to hear Dr. Starzl's comments on this possibility.

Dr. Starzl also suggests in his manuscript that FK might exert its effect by inhibiting an enzyme, PPIAse, which is a component of the receptor sites for both cyclosporine and FK. I would like to ask Dr. Starzl whether this enzyme is also a component of other major immunomodulating receptors on the lymphocyte surface, such as CD4, CD8, or DR, for example, and whether the expression of such other markers is affected in any way by FK.

My final question relates to the more profound immunosuppressive efficacy of FK. Could this be the 'missing link' in our life-long goal of inducing specific tolerance to allografts in humans?

Dr. G. Klintmalm (Dallas, Texas): I want to express my profound admiration for Dr. Starzl, not just for the work that he has presented, but also for bringing this drug from the laboratory to the clinical setting. This is truly a hallmark presentation, and I think we see here the first drug in the next generation of immunosuppressive drugs.

As indicated in the abstract, we do not see as impressive a result after kidney transplantation as with heart–lung or liver transplantation. The same has actually been reported by Dr. Starzl's group in the canine model. Do you believe there may be a first-pass effect of FK that may be responsible for this difference?

Dr. Oscar Salvatierra, Jr. (San Francisco, California): There is no group in the United States clinically using FK currently other than the Pittsburgh group, and they are to be commended for their courage and leadership role with this new agent.

Previous reports by the Pittsburgh group describe FK in rescue of failing grafts under cyclosporine therapy. This report now describes the first use of FK as the primary immunosuppressive agent in liver, kidney, heart, heart–lung and double lung transplantation from the time of graft implantation.

The report compares the use of FK with a good historical control group of patients treated with cyclosporine, the drug that heralded the new era of improved transplant success in the 1980s.

It appears from this report, and if the findings are confirmed in future clinical trials that will soon begin at other centers, that organ transplantation may now be entering another era of success with hopefully concomitant reduced sequelae from multiple drug therapy protocols that are currently in use.

Of great interest is the early significantly improved graft and patient survival rates in liver, heart, and/or lung transplantation and decreased retransplantation rates in these groups.

The kidney group is more difficult to evaluate, in that 10 of the 36 patients had other nonrenal organ transplants and nine had positive cytotoxic cross-matches against the donor, conditions that would have generally precluded renal transplantation at many centers. Yet 75% graft survival was achieved in this complex group.

The lower incidence of rejection episodes in the FK group of patients is extremely important, as it portrays a lesser need for increased or prolonged steroid and other drug usage. In fact, rejection was in general easily and successfully treated with just one pulse steroid bolus in most patients. More importantly, most patients were off steroids or on very low steroid dosages. Off steroids were 100% of the heart–lung and lung recipients, greater than one third of the liver recipients, and 71% of the kidney recipients.

If one examines these early findings as presented, the 1990s may well witness a new and meaningful breakthrough in immunosuppressive management.

I have two questions for Dr. Starzl. Have you established immunologic and outcome criteria by which you can safely convert patients to FK monotherapy, eliminating steroids?

Second, kidney recipients appear to require higher doses of FK to maintain equivalent plasma levels, as the liver recipients presumably related to the initial pass of the drug through the liver and its metabolism by that organ. How critical and important is the careful drug dosing by plasma levels in the kidney recipient, and how reliable is the currently used assay methodology?

Dr. Joel D. Cooper (St. Louis, Missouri): As has been pointed out many times, cyclosporine really ushered in an explosion of transplant activity, but we have all been hoping for something better. Perhaps FK is that next giant leap forward.

Primarily I wanted to thank him and acknowledge his assistance with one of our patients, a woman who had a double-lung transplant and did very well for 15 months, but who then went into fulminant and unrelenting rejection. With his encouragement and advice and his offer to help us with FK 506 on a compassionate basis, we replaced both her lungs. She is now out of the hospital and has done very well.

I realize that one mouse is no mouse, but certainly in this case the use of FK 506 greatly simplified the management of this patient. Dr. Starzl I owe you a piece of chocolate cake because she is a wonderful cook and brought me a chocolate cake the other day. I think I was supposed to bring you a piece, but I ate it.

Dr. Joseph E. Murray (Wellesley Hills, Massachusetts): It is challenging to learn of new drugs that are improving the results of human organ transplantation. It was in 1962 at a meeting of this Association that we first described the effectiveness of immunosuppressive drugs for renal transplantation in dogs and man (Ann Surg 1962; 156:337).

The drugs used then were azathioprine and actinomycin C. The use of steroids as a supplement soon followed. Cyclosporine came onto the scene about 15 years ago, and now we have FK 506.

I've always been interested in the mechanism of action of these various drugs, so I'd like Dr. Starzl's opinion as to whether or not all drugs act differently or is there a final common pathway of action. Tom has had wide experience going back to the early days of Imuran treatment.

In our canine renal transplants in the mid 1960s we were pleased and surprised to be able to wean completely some of the long-term allografted dogs from the drugs. The same happened in an occasional patient in whom we were forced to withdraw therapy for medical reasons. Do you feel that FK506 may be able to produce a permaent tolerance? Is it easier to withdraw therapy using FK506 than with other drugs?

Dr. Robert E. Condon (Milwaukee, Wisconsin): I would like to ask a few questions that don't have to do with the use of this drug as an

immunosuppressant but relate to its status as a macrolide antibiotic. The first has to do with possible effects on gut motility. The macrolides stimulate gut smooth muscle in various ways. That is what accounts for the nausea that is occasionally associated with the administration of erythromycin, for example.

So my question is: In your patients, have you noted any change in bowel habits, and if so, is the change in transit time responsible for the changes that you have noted in the cholesterol levels in your patients?

The second question has to do with possible antibacterial effects of this macrolide. There was a startling improvement in the overall sepsis rate in these patients and, although most of that is probably due to the fact that they got less steroids, could it possibly be related to some direct antibacterial effect of the macrolide drug?

PROFESSOR PETER J. MORRIS (Oxford, England): I would like to ask Dr. Starzl to comment on the histology. I was very excited about 6 months ago when I saw some histology from routine biopsies in patients with liver transplants at about 2 to 3 weeks after the transplant, showing no evidence of rejection whatsoever. This was a fairly unique finding in the relatively small number of transplants done at that time.

I gather that cardiac transplants on routine biopsy do show a cellular infiltrate that looks not dissimilar from what is seen with cyclosporine. Again that was, of course, in the first half-dozen patients or so, and I would like a follow-up on that. Also, I would be interested to know if there is any information on the histology of the kidney transplants. Certainly the histologic findings in the liver transplants were absolutely dramatic and I just wonder if that is unique to the livers.

DR. THOMAS E. STARZL (Closing discussion): I particularly wanted to say hello and thank you to Dr. Murray, to tell him how pleased I am to have him hear this paper and to acknowledge the fact that the first steps toward the summit, which he took, were the most important ones. The question of tolerance induction that Felix Rapaport already discussed is a very difficult one. I think as you (Dr. Murray) yourself showed and we have emphasized over the years, the accidental achievement of tolerance, if that is what it is, probably follows the same pathway using all drugs, and so what you saw with those early Boston dogs named Connecticut and Massachusetts is probably what we can see today, and induce with FK 506 but apparently with greater predictability than at any time in the past. It is very difficult to get papers published that actually examine the mechanism of graft acceptance, because all such papers interdict the opinions or biases of most of the referees. We have such a manuscript out there at the moment, which suggests that all hypotheses up to the present are wrong. The truth is that we do not know why some grafts are accepted, and others are not.

Of all of the discussions of this morning, I was actually most intrigued and learned the most from that of Dr. Condon because he is an expert on infectious disease. If Dick Simmons is here, I hope that he heard Dr. Condon's two questions, the first being about the potential antibiotic effect of the FK. This is a fascinating suggestion, and I wish I had thought of it myself. We will follow up your idea, and if you are right, we will claim it as our own.

Concerning the second question, I think that there potentially is an effect on GI motility and GI function, although it is very hard to detect. In dogs, the GI toxicity of FK 506 was actually what scared off the Cambridge group because it made the dogs so sick that many of the animals developed intussusception. This was the toxicity that we had feared the most in our first humans, but it did not materialize.

Dr. Klintmalm, I suspect that there is a first-pass removal of FK 506 by the liver and this may make it easier to treat liver recipients who take the drug by mouth and who therefore have it presented in high concentration to the liver. Because a diminished amount leaks through, this may make it necessary for larger doses to be given for organs not exposed to the splanchnic venous return. If this is a genuine dose-adjustment phenomenon, it must also apply, although it has never been studied formally, with cyclosporine. Many of the kinetics including liver metabolism of cyclosporine and FK 506 are similar. So far, the bottom line is that the liver recipients do require a smaller dose, as discussed in the manuscript.

Dr. Salvatierra also has asked important questions, of which the foremost is which patients can have the steroids stopped. I wish we had a foolproof formula for prediction. What we do in practice is go down to zero steroids automatically, and then if we get stung with a rejection, we resume steroids. The implications of a rejection under those circumstances are less grave than in the past, since the patients usually can be rescued rather easily from such rejections. Thus, the game is worth the candle to try steroid discontinuance at least once with the liver recipients.

To answer Peter Morris' question about the liver biopsies, there is evidence of rejection in many of the biopsies. In kidney grafts, the same is true. Drs. Jake Demetris and Barbara Banner, our pathologists, did a blind study of cyclosporine *versus* FK 506-treated kidneys. There was a conviction at one time that the vascular lesions of rejection would be more common in the FK patients. This was not the case, and in fact the findings were the other way around. The vascular lesions occurred at about one-third the rate in the FK 506 kidneys compared with the conventional cyclosporine regimen. It is difficult with blind reading to make a qualitative distinction about rejection patterns in any given case. In both kidney and liver grafts, eosinophils may be prominent with either drug. I think that this occurs when there is an effort to move downward in steroid doses.

The heart biopsies are very difficult to interpret. These biopsies tend to guide therapy, since there is not a biochemical litmus paper as we have with the other organs. The dependence on cardiac graft biopsies is very nearly complete, but whether or not those cells that are being found in the endomyocardial biopsies should actually trigger responses in therapy is an open question. What we are doing in many of our patients whose grafts do have these cells, is to make minimal and sometimes no therapeutic adjustment if the patients are well clinically.

Felix Rapaport and some one else also asked, what can be done with FK 506? Can it play a role in research in heterotransplantation? That, of course, is the next step, and I think the answer is yes, but. The 'but' is that something has to be done about the heterospecific preformed antibodies that are present with cross-species grafting. Twenty-seven years ago, we showed how close to possible it was even then to transplant baboon kidneys to humans. I believe that the cell-mediated rejections that occurred with those baboon-to-human transplantations could be controlled with the drugs we have today, but what we can not control are the humoral rejections. So, Felix, I think that this is the component of the problem that we must work on. We are advising against such trials, at least at the moment.

I see that the warning light is blinking that precludes my answering the most important question of all about the peptidyl-prolyl isomerase (PPIase) enzyme system. This enzyme is contained in the binding sites of both cyclosporine (cyclophilin) and FK 506 (binding site not yet named). These are distinct binding sites, and yet they have as their principal constituent the PPIase, which was discovered 5 years ago. PPIase had no known function until about a year ago, when it was recognized to be the principal constituent of cyclophilin. Now it is assumed that inhibition of PPIase is central to the action of both FK 506 and cyclosporine, even though FK 506 has no biochemical relationship to cyclosporine. What has happened here is that by accident a novel new class of cytosolic proteins (binding sites) has been discovered that is involved across a broad range of signal transduction processes that control the interior chemistry of the cell, not limited to immune modulation. This has been evident by studying the effects of the two PPIase inhibitors (cyclosporine and FK 506) on numerous physiologic processes, including carbohydrate, cholesterol, and uric acid metabolism; control of liver regeneration (both drugs augment this process), neurologic functions, and hair growth, to supply an incomplete list.

Finally, I want to pay tribute to Joel Cooper, the courageous Washington University surgeon who earlier today gave a classic paper on lung transplantation. The case he mentioned in discussing our paper is the first successful double lung retransplantation of which I am aware. It must have been a technical *tour de force*, and we were proud to help him from a distance.