SYNOPSIS

Name of Sponsor/Company: Astellas Pharma Europe Ltd.		
Name of Finished Product: FK506 (Prograf)		
Name of Active Ingredient: Tacrolimus		

Title of Study: An Open, Randomized, Multicenter, Clinical Study to Compare the Safety and Efficacy of Tacrolimus and Minimal Steroids in Combination with Either a Monoclonal Anti-IL2R Antibody (Daclizumab) or Mycophenolate Mofetil in Liver Allograft Transplantation

Responsible Medical Officer/Coordinating Investigator: [redacted]
Astellas Pharma Europe Ltd., [redacted]

Investigator(s): [redacted]

Study Center(s): 37 centers participated in the study. Belgium (3), Finland (1), Germany (8), Great Britain (1), Hungary (1), Italy (8), Poland (2), Romania (1), Spain (9), Sweden (1), Switzerland (2).

Publication (reference): None available to date.

Study Period: **Date of First Enrollment:** 15 March 2005 **Date of Last Evaluation:** 22 June 2007	**Phase of Development:** Phase III

Objectives: The objective of this study was to evaluate the efficacy and safety of two regimens containing tacrolimus and minimal steroids together with daclizumab or mycophenolate mofetil (MMF).

Study Design: This was a multicenter, randomized, open, two-arm, parallel group, comparative study.
Group 1: Tacrolimus + daclizumab + i.v. steroid bolus peri-operatively [TAC/DAC].
Group 2: Tacrolimus + MMF + i.v. steroid bolus peri-operatively [TAC/MMF].

Patients were randomized 1:1 to treatment groups with stratification by study center and HCV status.

Diagnosis and Main Criteria for Inclusion:
Male or female patients with minimum 18 years of age and were suitable candidates for an orthotopic liver or split liver transplant were eligible for the study. Patients who received a liver transplant from a deceased or living donor and who provided informed consent were included.

Number of Subjects (planned and analyzed): Based on a (two-sided) nonparametric comparison of the two groups at level α, 255 patients per treatment arm (550 patients in total) were needed to detect a difference of 10% at Month 3 with a power of at least 80%. Assuming a drop out rate of 10%, 600 patients were to be randomized, 300 per treatment group. In total, 627 patients were randomized. The full analysis set (FAS) consisted of 305 patients in the TAC/DAC group and 297 patients in the TAC/MMF group.

Test Product, Dose And Mode of Administration:
The dosing of tacrolimus was the same for both groups. The initial daily dose was 0.10mg/kg to 0.15mg/kg p.o. Subsequent oral tacrolimus doses were to be adjusted on the basis of clinical evidence of efficacy and occurrence of adverse events and using the following recommended whole blood trough level ranges: Day 0–42, 10 to 15ng/mL; Day 43–93, <10ng/mL.

Lot Numbers: Tacrolimus 0.5mg: [redacted]; Tacrolimus 1.0mg: [redacted]; Tacrolimus 5.0mg: [redacted]

[redacted].

Duration of Study and Treatment: Patients were followed for 3 months with 7 scheduled assessment visits.

Criteria for Evaluation: The primary efficacy endpoint was the incidence of and time to first biopsy-proven acute rejection (BPAR) which required treatment within 3 months following transplantation. Secondary efficacy and safety endpoints were: overall incidence of acute rejection; incidence of and time to first acute rejection; incidence of and time to first corticosteroid-resistant acute rejection; Overall frequency of BPAR; incidence of and time to first BPAR; incidence of and time to first steroid resistant BPAR; severity of BPAR; patient and graft survival; renal function (assessed by calculated creatinine clearance); incidence of adverse events; absolute change in serum lipids; incidence of diabetes mellitus; incidence of hypertension; occurrence of CMV infection.

Statistical Methods:

Primary efficacy analysis: The primary end point was the incidence of and time to first biopsy proven acute rejection which required treatment within 3 months of transplantation. The two treatment arms were compared and tested for differences of survival functions of all patients of the FAS.

The null and alternative hypotheses were: H_0: $S_{Daclizumab}(t) = S_{MMF}(t)$ against H_a: $S_{Daclizumab}(t) \neq S_{MMF}(t)$where $S_{Daclizumab}(t)$ and $S_{MMF}(t)$ denoted the survival function for the TAC/DAC arm and TAC/MMF arm, respectively.

H_0 was to be rejected if the p-value of the two sided Wilcoxon-Gehan test was below 0.05. Analysis of the primary endpoint was also performed stratified by patients with positive or negative HCV status.

Secondary efficacy and safety analyses: The incidence and time to first acute rejection, corticosteroid-resistant acute rejection, BPAR and steroid resistant BPAR were analysed using Kaplan-Meier cumulative survival probability estimates. The difference between survival estimates was assessed using the Wilcoxon-Gehan test (two sided). The 95% confidence interval (Greenwood formula for SD and normal approximation) was calculated for the difference in survival at 3 months. The number of patients who experienced an AR or a BPAR was compared between treatment arms with the two sided chi-squared (Fisher's exact) test. Banff criteria were used to classify the severity of a BPAR.

Patient and graft survival were analysed using Kaplan-Meier cumulative survival probability estimates. The difference between treatment groups was assessed using the Wilcoxon-Gehan test. The 95% confidence intervals (Greenwood formula for SD and normal approximation) were calculated for the difference at 3 months.

Secondary efficacy and safety analyses: All secondary endpoints were summarized per treatment group using appropriate descriptive statistics, i.e.: number (%) of patients for categorical variables, and mean, standard deviation, median, minimum, maximum for continuous variables.

Treatments were compared with chi-squared or Fisher's exact test. Time to event was analyzed using Kaplan-Meier cumulative survival probability estimates and comparisons of treatments were analyzed using the Wilcoxon-Gehan test.

Analysis sets: The primary analysis of efficacy data was based on the FAS defined as all randomized patients who received at least one dose of study medication and were transplanted with results attributed to the treatment group to which they were randomized. All analyses were based on the FAS.

RESULTS:

Analysis Sets and Subject Disposition:

There were 627 patients enrolled in the study and randomized to treatment. The population available for analysis is summarized in the table below.

Table 1: Populations for Analysis – Number of Patients

	TAC/DAC	TAC/MMF	Total
Randomized	315	312	627
Excluded from full analysis set	10	15	25
Not transplanted, no study medication	8	9	17
Not transplanted, study medication	2	6	8
Full analysis set (FAS)*	305	297	602

* All randomized and transplanted patients who received at least one dose of study medication (tacrolimus, daclizumab, or MMF).

The FAS consisted of 305 patients in the TAC/DAC group and 297 patients in the TAC/MMF group. All analyses were based on the FAS.

Approximately 70% of the randomized patients in each group completed the study, with the most common reason for withdrawal being due to an adverse event for both treatment arms. Thirty-one patients died during the study and 19 patients died following study withdrawal. The causes of patient death during the study in the TAC/DAC group were sepsis/multiorgan failure (1.6%), cardiac failure/cardiac arrest (1.3%), hemorrhage (1.0%), and pericardial effusion, pneumonia, and respiratory failure (all 0.3%). The causes of patient death during the study in the TAC/MMF group were sepsis/multiorgan failure (4.0%), and cardiac failure/cardiac arrest, graft failure, pontine myolinolysis, pulmonary embolism (all 0.3%). Sepsis/multiorgan failure was also the most common cause of death after study withdrawal in both groups (2.3%, TAC/DAC; 2.7%, TAC/MMF).

Table 2: Patient Disposition - Number (%) of Patients

	TAC/DAC N=305	TAC/MMF N=297	Total N=602
Completed	220 (72.1)	204 (68.7)	424 (70.4)
Total deaths	25 (8.2)	25 (8.4)	50 (8.3)
During study	15 (4.9)	16 (5.4)	31 (5.1)
After withdrawal/EOS	10 (3.3)	9 (3.0)	19 (3.2)
Withdrawn*	70 (23.0)	77 (25.9)	147 (24.4)
Retransplantation	13 (4.3)	13 (4.4)	26 (4.3)
Adverse event	45 (14.8)	46 (15.5)	91 (15.1)
Lost to follow-up	1 (0.3)	2 (0.7)	3 (0.5)
Protocol violation	6 (2.0)	12 (4.0)	18 (3.0)
Other	5 (1.6)	4 (1.3)	9 (1.5)

FAS

* For reasons other than death.

Demographics:

Patients in the two treatment groups were well-matched on demographics and baseline characteristics; in particular, there were no differences associated with a p-value of <0.05. There were twice as many males as females in both groups (approximately 68% and 32%). The mean age was comparable at 53.7 years (±9.4) in the TAC/DAC group and 53.1 years (±9.5) in the TAC/MMF group.

The majority of patients in both groups presented with a primary diagnosis of cirrhosis (67.0%,

TAC/DAC; 70.0%, TAC/MMF). Carcinoma was the second most common primary diagnosis in both groups (23.3% and 18.9%, respectively). Approximately 73% of patients in both groups were CMV positive at baseline.

Donor demographics, donor/recipient ABO, donor type, and total ischemia time were comparable for the two treatment groups. The number of CMV negative recipients receiving an organ from a CMV positive donor was similar (-recipient/+donor): 12.8%, TAC/DAC; 14.8%, TAC/MMF.

Study Drug Exposure:

Recommended tacrolimus blood trough levels were 10–15ng/mL between Days 0 and 42 and <10ng/mL from Day 43 to end of study for both groups: mean tacrolimus trough levels were slightly above recommended ranges in the TAC/DAC group at Month 2 but decreased to within the recommended range by month 3/end of study. Mean tacrolimus trough levels were within recommended ranges during all assessment periods in the TAC/MMF group. At Month 3, the mean tacrolimus trough levels were comparable and within targeted ranges in both groups: 9.22ng/mL (±3.34) in the TAC/DAC group and 9.44ng/mL (±3.80) in the TAC/MMF group (for completers).

Primary Endpoint and Efficacy Results:

There was no evidence of a difference between treatments in the incidence of or time to first BPAR requiring treatment within 3 months. The incidence of BPAR requiring treatment was 16.4% (50 patients) in the TAC/DAC group compared with 15.5% (46 patients) in the TAC/MMF group. At Month 3 the Kaplan-Meier estimate for the rate of patients free from BPAR requiring treatment was 0.815 in the TAC/DAC group (95% confidence interval: 0.768 to 0.861) and 0.822 in the TAC/MMF group (95% confidence interval: 0.773 to 0.871). The Kaplan-Meier estimate for the corresponding difference in treatment groups was -0.007 (95% confidence interval: -0.075 to 0.060).

Table 3: Overall Estimated Rate of Patients Free From Biopsy-proven Acute Rejection requiring Treatment (Kaplan-Meier Method)

	TAC/DAC N=305			**TAC/MMF N=297**		
	Number of Patients		Rejection-free rate*	Number of Patients		Rejection-free rate*
	With events	Remaining at risk		With events	Remaining at risk	
Week 1	15	268	0.948	22	254	0.922
Week 2	13	243	0.902	15	220	0.865
Week 3	3	236	0.890	5	201	0.845
Week 4	6	220	0.867	2	195	0.837
Month 2	12	196	0.819	1	175	0.832
Month 3	1	68	0.815	1	63	0.822

FAS

*Rate of patients free from biopsy-proven acute rejection which required treatment.

An analysis of the primary endpoint, incidence of or time to first BPAR requiring treatment, was performed on patients who were either HCV negative or HCV positive at baseline. This analysis did not reveal differences between treatment groups. The incidence of BPAR requiring treatment in HCV negative patients in the TAC/DAC group was 18.8% (39/208 patients) compared with 15.0% (31/207 patients) in the TAC/MMF group. In patients who were HCV positive, the incidence of BPAR requiring treatment was 11.7% (11/94 patients) in the TAC/DAC group compared with 16.9% (15/89 patients) in the TAC/MMF group.

In both treatment groups, more BPAR events occurred during the first two weeks of the study than at other times. During the first two study weeks a BPAR was reported in 28 patients in the TAC/DAC

group and in 37 patients in the TAC/MMF group. However, after Week 2 to end of study, more than twice as many patients in the TAC/DAC group than in the TAC/MMF group experienced a BPAR requiring treatment: 22 patients vs. 9 patients, respectively.

The number of patients in each treatment group experiencing an acute rejection was similar as presented in Table 4 below:

Table 4: Overall Frequency of Acute Rejections

	TAC/DAC N=305		**TAC/MMF N=297**	
	Patients n (%)	Episodes	Patients n (%)	Episodes
Acute rejection	74 (24.3)	81	65 (21.9)	70
Spontaneously resolving	22 (7.2)	23	16 (5.4)	16
Corticosteroid sensitive	40 (13.1)	41	44 (14.8)	45
Corticosteroid resistant	12 (3.9)	12	8 (2.7)	8
Resolved with treatment	1 (0.3)	1	2 (0.7)	2
Unresolved with treatment	1 (0.3)	1	0 (0.0)	0
Unresolved without treatment	10 (3.3)	10	6 (2.0)	6
Other	5 (1.6)	5	1 (0.3)	1

FAS

There was no statically significant difference in the estimated graft survival rates as estimated by the Kaplan-Meier method. The estimated rate of graft survival was 88.2% in the TAC/DAC group and 87.8% in the TAC/MMF group. The incidence of graft loss during the study was equivalent in the two groups at 9.2% in the TAC/DAC group and 9.8% in the TAC/MMF group. Incidences of graft loss following study withdrawal or at end of study were also similar in the treatment groups at 2.6% and 2.4%, respectively.

Safety Results:

Patient Survival

The estimated rate of patient survival was similar at 91.8% in the TAC/DAC group and 91.6% in the TAC/MMF group.

Adverse Events

As expected in this patient population, almost all patients in both treatment groups experienced at least one adverse event (AE); 95.7% in the TAC/DAC group and 97.6% in the TAC/MMF group, differences were not significant. The incidence of bacterial infections and blood and lymphatic system disorders (in particular leukopenia) was significantly higher in the TAC/MMF group while the incidence of headache and cardiac disorders (in particular supraventricular arrhythmias) was higher in the TAC/DAC group.

Table 5: Incidence of the Most Frequently Reported* Adverse Events Regardless of Relationship to Study Medication – Number (%) of Patients, Events

MedDRA SOC High Level Term *Preferred Term*	**TAC/DAC N=305**		**TAC/MMF N=297**	
	Patients	Events	Patients	Events
Infections/Infestations	**159 (52.1)**	**102**	**166 (55.9)**	**288**
Bacterial †	10 (3.3)	10	25 (8.4)	28
Urinary tract infection bacterial	27 (8.9)	28	23 (7.7)	23
Pneumonia bacterial	18 (5.9)	18	20 (6.7)	21
Cholangitis supurative	19 (6.2)	19	18 (6.1)	18
Cytomegalovirus	19 (6.2)	19	24 (8.1)	24
Hepatitis C	22 (7.2)	22	15 (5.1)	15
Sepsis, bacteremia, viremia	19 (6.2)	20	18 (6.1)	18
Abdominal/gastrointestinal	17 (5.6)	18	18 (6.1)	21
Renal/Urinary disorders	**133 (41.6)**	**151**	**117 (39.4)**	**133**
Renal failure	80 (26.2)	87	66 (22.2)	74
Renal impairment	37 (12.1)	44	28 (9.4)	28
Renal failure acute	14 (4.6)	14	20 (6.7)	20
Metabolism/Nutrition disorders	**111 (36.4)**	**151**	**126 (42.4)**	**164**
Hyperglycemia	55 (18.0)	55	57 (19.2)	59
Hypoalbumemia	20 (6.6)	20	24 (8.1)	24
Hyperkalemia	17 (5.6)	17	21 (7.1)	22
Diabetes mellitus	19 (6.2)	19	20 (6.7)	21
Gastrointestinal disorders	**100 (32.8)**	**171**	**116 (39.1)**	**182**
Diarrhea	46 (15.1)	50	61 (20.5)	71
Ascites	17 (5.6)	17	20 (6.7)	20
Nausea/vomiting	21 (6.9)	28	16 (5.4)	21
Hepatobiliary disorders	**97 (31.8)**	**136**	**103 (34.7)**	**121**
Cholestasis	57 (18.7)	60	58 (19.5)	59
Bile duct stenosis	16 (5.2)	16	8 (2.7)	9
Hepatic vascular disorders	14 (4.6)	15	20 (6.7)	23
Blood/Lymphatic system disorders‡	**77 (25.2)**	**104**	**110 (37.0)**	**157**
Anemia	48 (15.7)	50	62 (20.9)	64
Thrombocytopenia	21 (6.9)	22	34 (11.4)	37
Leukopenia§	13 (4.3)	15	31 (10.4)	34
Respiratory/Thoracic/Mediastinal disorders	**79 (25.9)**	**103**	**81 (27.3)**	**116**
Pleural effusion	47 (15.4)	50	55 (18.5)	62
Injury/Poisoning/Procedural complications	**72 (23.6)**	**94**	**77 (25.9)**	**93**
Complications of transplanted liver	20 (6.6)	20	24 (8.1)	24
Investigations	**75 (24.6)**	**106**	**65 (21.9)**	**93**
Liver function analyses	51 (16.7)	57	35 (11.8)	42
Hepatic enzyme increased	29 (9.5)	32	19 (6.4)	20
Renal function analyses	23 (7.5)	24	20 (6.7)	21
Blood creatinine increased	22 (7.2)	23	20 (6.7)	21
Vascular disorders	**72 (23.6)**	**78**	**60 (20.2)**	**65**
Hypertension	47 (15.4)	48	38 (12.8)	38
Nervous system disorders	**59 (19.3)**	**78**	**69 (23.2)**	**87**
Tremor	19 (6.2)	20	28 (9.4)	29
Headache¶	18 (5.9)	18	6 (2.0)	6
General disorders/Administration site conditions	**42 (13.8)**	**52**	**53 (16.8)**	**60**
Pyrexia	14 (4.6)	16	21 (7.1)	22
Peripheral edema	13 (4.3)	13	17 (5.7)	17

Psychiatric disorders	**31 (10.2**	**38**	**35 (11.8)**	**47**
Cardiac disorders**	**43 (14.1)**	**56**	**21 (7.1)**	**24**
Supraventricular arrhythmias††	19 (6.2)	21	8 (2.7)	8
Musculoskeletal/Connective tissue disorders	**21 (6.9)**	**23**	**25 (8.4)**	**31**
Back pain	8 (2.6)	8	17 (5.7)	19
Skin/Subcutaneous tissue disorders	**20 (6.6)**	**20**	**15 (5.1)**	**16**

FAS

SOC=System Organ Class. Adverse events were coded using Version 8.0 of MedDRA. Adverse events coded as liver transplant rejection, transplant rejection are not included. *AE occurred in at least 5% of patients in either treatment group. †p=0.008; ‡p=0.002; §p=0.004; ¶p=0.02; **p=0.005; ††p=0.048; Fisher's exact test for all comparisons.

There was a significant difference in the incidence of causally-related adverse events: 76.1% incidence in the TAC/DAC group vs. an incidence of 82.8% in the TAC/MMF group (p=0.04, Fisher's exact test). AEs which occurred significantly more often in the TAC/MMF group than in the TAC/DAC group were diarrhea (p=0.003) and anemia (p=0.001), and leukopenia and thrombocytopenia (p< 0.001) (Fisher's exact test for all comparisons).

A comparable number of patients in each treatment group died as a result of an adverse event (15 patients, TAC/DAC; 14 patients, TAC/MMF). The most commonly reported adverse event leading to patient death in both treatment groups was infection which was the cause of death in 6 patients in the TAC/DAC group and in 8 patients in the TAC/MMF group. All adverse event-related causes of death were of the type expected in recipients of a liver transplant. There were no significant differences between the groups in any of the adverse events causing patient death as shown in the following table:

Table 7: Adverse Events Leading to Death – Number (%) of Patients, Events

MedDRA primary SOC	**TAC/DAC N=305**		**TAC/MMF N=297**	
	Patients	Events	Patients	Events
Infections/infestations	**6 (2.0)**	**7**	**8 (2.7)**	**8**
General disorders/administration site conditions	**3 (1.0)**	**3**	**4 (1.3)**	**4**
Cardiac disorders	**4 (1.3)**	**4**	**1 (0.3)**	**1**
Injury/poisoning/procedural complications	**2 (0.7)**	**2**	**1 (0.3)**	**1**

FAS; SOC=System Organ Class. Adverse events were coded using Version 8.0 of MedDRA.

The incidence of serious causally-related adverse events was comparable at 28.9% in the TAC/DAC group and 30.6% in the TAC/MMF group. The incidence of types of events was comparable between groups except for blood and lymphatic system disorders which were significantly more frequent in the TAC/MMF group than in the TAC/DAC group (p=0.005), and cardiac disorders which were significantly more frequent in the TAC/DAC group than in the TAC/MMF group (p=0.038) (Fisher's exact test for all comparisons).

The incidence of premature study discontinuation due to an adverse event was similar at 23.6% in the TAC/DAC group and 24.9% in the TAC/MMF group. Premature study discontinuation was most commonly due to a nervous system disorder in both groups: 3.9%, TAC/DAC and 6.1%, TAC/MMF. Significantly more patients in the TAC/DAC were prematurely withdrawn from the study due to immune system disorders (liver transplant rejection) (p=0.007) and significantly more patients were withdrawn due to blood and lymphatic system disorders (leukopenia) (p=0.029) in the TAC/MMF group (Fisher's exact test for all comparisons).

The incidence of *de novo* diabetes mellitus (at least 2 fasting glucose values ≥7.0mmol/L or >30 days antidiabetic treatment) from month 2 to month 3 was similar in the two groups at 9.5%, TAC/DAC and 11.0%, TAC/MMF with approximately 3.3% of patients in both groups receiving any type of antidiabetic medication at end of study.

No relevant differences were observed between treatment groups in serum creatinine, creatinine clearance, and glomerular filtration rate (GFR) mean values. Similarly, no differences were noted in hepatic function or in serum lipid values. Treatment groups were also comparable with respect to other laboratory parameters, vital signs, and mean length of hospitalization.

CONCLUSIONS:

This randomized clinical trial has demonstrated that excellent results may be achieved in liver transplantation using a tacrolimus-based immunosuppressive regimen which avoids the use of steroids other than as a one-time bolus dose given peri-operatively. Comparable outcomes, in terms of frequency of rejection, patient and graft survival, and incidences of adverse events, were achieved when either daclizumab or MMF was combined with tacrolimus.

Date of Report: June 2008