EXECUTIVE SUMMARY
NDA 50-777
PROTOPIC (0.03% and 0.1% Tacrolimus [FK506]) OINTMENT

1. Introduction

The active ingredient in Protopic ointment is Tacrolimus (FK506), an immuno-suppressant macrolide that has been approved as Prograf capsules (NDA 50-708) and injection (NDA 50-709) for the prophylaxis of organ rejection in patients receiving allogeneic liver transplants. In the present NDA, the sponsor proposes the use of ointment formulations for the short and long term treatment of the signs and symptoms of atopic dermatitis in adult and pediatric patients two years of age or older. Protopic ointments, 0.03% and 0.1% will be applied topically twice daily as a thin layer to affected areas of skin. Other related drugs are Cyclosporin A and Rapamycin. Cyclosporin A (Neoral) has been approved for the oral treatment of recalcitrant psoriasis.

2. Clinical Studies

2.1 Protocols

Twenty eight clinical studies were submitted in the present NDA. Eleven of these are Clinical pharmacology/pharmacokinetic studies including the topical safety patch tests. The remaining 17 studies consisted of 5 pivotal phase 3 studies and 12 supportive phase 2 and phase 3 studies. Efficacy evaluation depends primarily on the adequate and well-controlled pivotal studies, whereas safety evaluation includes the full global experience.

The five pivotal studies consist of:

1) Three similarly designed, placebo controlled, randomized, double blind and multi-center studies. These studies were powered to detect differences in efficacy and safety between the active and vehicle arms. Each study consisted of three arms: vehicle, 0.03% and 0.1% tacrolimus ointments. Treatments were for 12 weeks. The enrolled patients had moderate to severe atopic dermatitis involving at least 10% of the body surface area. Two of these studies were in the adult population, and one was in the pediatric population (ages 2 to 15 years).
2) Two open-label long-term safety studies. Both studies used 0.1% tacrolimus ointment treatments for up to one year. One study was in the adult population, and the other was in the pediatric population.

There was no comparison with topical steroids in these pivotal studies. The sponsor has submitted other clinical studies comparing tacrolimus ointment with topical steroids. However, these studies were not placebo controlled.

2.2 Efficacy

Efficacy evaluation depends mainly on the placebo controlled pivotal studies. The primary parameter was the rate of success defined as a rating of cleared or excellent improvement in the physician's global evaluation, i.e., ≥90% improvement in the areas defined by the investigator as active disease for treatment at baseline. For the Physician's Global, changes in the overall status of the atopic dermatitis lesions identified for treatment at baseline were rated using the following scale:

Cleared	100%,	Excellent Improvement	90-99%,
Marked Improvement	75-89%,	Moderate Improvement	50-74%,
Slight Improvement	30-49%	No Appreciable Improvement	0-29%,
Worse	< 0%.		

A- ADULT STUDIES:

For the primary efficacy parameter, a significantly greater success rate was observed for each tacrolimus ointment treatment group compared with the vehicle in each study and in both studies combined.

In addition, the combined two adult studies (probably as a result of increase in power obtained from pooling), as well as the results of one adult study (#036) showed a statistically significantly greater success rate for the 0.1% tacrolimus ointment treatment group compared with the 0.03% tacrolimus ointment treatment group.

Analysis of success in different sub-populations showed that the 0.1% formulation is significantly more effective than the 0.03% formulation in the following sub-populations of patients: females, blacks, severe disease and extensive disease involvement (>75% BSA).

B- PEDIATRIC STUDY:

A significantly greater success rate was observed for each tacrolimus ointment treatment group compared with the vehicle.

However, the results of this study failed to show any statistically significant greater success rate for the 0.1% tacrolimus ointment treatment group compared with the 0.03% tacrolimus ointment treatment group, although a trend can be observed (1.5 – 4.0% difference, depending on the population studied). Detailed efficacy analysis of the pediatric study is shown in Appendix 1.

The sponsor claims a statistically significant difference in the time to first at least slight improvement, being significantly shorter (p=0.039) in the 0.1% tacrolimus group compared with the 0.03% tacrolimus group (Appendix 2). This parameter was not a previously defined efficacy endpoint in this study. However, the time to first excellent, marked or moderate improvement was comparable between the two tacrolimus treatment groups. Also, the data (Appendix 2) show that this statistically significant difference in the mean time to first slight improvement is only 0.7/11.6 days, a clinically insignificant difference.

Analysis of success in different sub-populations showed that the 0.1% formulation is not significantly better than the 0.03% formulation in any of sub-populations tested.

The sponsor has failed to demonstrate any superior efficacy of the 0.1% ointment vs. the 0.03% in the pediatric population. It was previously mutually agreed upon, in the end-of-phase 2 meeting of October 1996, that "the dose selected will be 0.03% unless 0.1% is significantly superior to 0.03%."

2.3 Safety

A- Pivotal Controlled Studies:

A total of 983 patients were treated in the three pivotal 12-week studies. Of these, 352 were pediatric patients (2-15 years old). A total of 55.2% of the patients were female, and the mean age was 27 years (range: 2 to 79 years of age). The majority of patients were white; however, blacks were well represented, comprising 26.6% of the patients. Atopic dermatitis affected >50% of the total body surface area at baseline in 40.9% of patients. The atopic dermatitis was severe in 58.1% of patients.

The common adverse events (5% incidence in any treatment group) in the three pivotal studies combined included flu-like symptoms, fever, allergic reaction, headache, increased cough, asthma, pruritus, the sensation of skin burning, skin erythema and skin infection. Generally, local irritation events were mild or moderate in severity and tended to be more common in patients with severe atopic dermatitis at baseline, or with >75% of their body surface area affected at baseline, and tended to be worse at the beginning of treatment.

Comparison of 12-week incidence rates, after adjustment for higher premature discontinuations in the vehicle arm, showed a statistically significant difference between one or both tacrolimus ointment treatment groups and vehicle in the following adverse events: the sensation of skin burning, pruritus, flu-like symptoms, headache, skin tingling, acne, folliculitis, alcohol intolerance, hyperesthesia, dyspepsia, myalgia, herpes zoster *(5 of 6 cases were chicken pox)*, and cyst.

A total of 16 (1.6%) patients (7 patients in the pediatric study and 9 patients in the adult studies) experienced one or more serious adverse events during treatment. The incidence of serious adverse events during treatment was similar for vehicle-treated (1.5%) and tacrolimus ointment-treated (1.7%) patients. In addition, seven patients (4 vehicle and 3 tacrolimus ointment-treated patients) experienced a serious adverse event post-treatment. There were no patient deaths in these studies.

In the three pivotal studies, a total of 68 patients had adverse events that led to discontinuation from the study. The more common adverse events leading to discontinuation were pruritus, skin erythema, the sensation of skin burning, and skin infection. A higher percentage of vehicle-treated patients than tacrolimus ointment-treated patients discontinued due to an adverse event.

B- Pivotal Long-Term Safety Studies:

Safety results of these studies were essentially similar to those of the three pivotal controlled studies. However, the daily hazard rate analysis of the combined controlled and uncontrolled (long-term) studies revealed a progressively increasing rate of lymphadenopathy through the three consecutive periods (3 months each) analyzed. Review of these cases of lymphadenopathy showed that most of them were lymphadenitis secondary to infections such as tonsillitis or skin infection. However, a few of them were unexplained or uninvestigated enlargement lymph

nodes. The clinical significance of this unexplained lymphadenopathy is unknown and is potentially of concern, especially when considered together with the preclinical toxicology studies showing high incidence of lymphomas in mice.

C- Global Experience:

Safety results from the global experience, including a total of 28 clinical studies, were essentially similar to the safety results of the 5 pivotal studies. In the global experience, the total number of lymphadenopathy cases was 23/3446 (=0.7%, Table 8.4.13.10.3.1 of NDA).

3. Potential Risks

In assessing the potential risks associated with topical tacrolimus therapy, the possibility of systemic toxicity associated with percutaneous penetration must be considered. Clinical pharmacokinetics studies conducted in this NDA have demonstrated that systemic levels of tacrolimus appear following topical application of tacrolimus ointment to atopic dermatitis skin. Recommended whole blood trough concentrations of tacrolimus for pediatric or adult liver transplant patients to achieve immunosuppression are 5-20 ng/ml. In the clinical studies of adults with atopic dermatitis, blood levels $\geq$ 5 ng/ml were detected in 1.6% of patients at some timepoint during their study enrollment; in the clinical studies of pediatric patients with atopic dermatitis, one 3-year old had a systemic blood concentration of 9.58 ng/ml of tacrolimus. Immune function tests in adults enrolled in the clinical studies revealed no defect in delayed type hypersensitivity following chronic exposure to topical tacrolimus. Immune function tests have not been conducted in pediatric patients following chronic exposure to topical tacrolimus.

Many of the toxicities associated with systemic tacrolimus are readily reversible upon discontinuation of systemic administration and/or were not observed in the clinical trials conducted in this NDA. Squamous cell carcinomas of the skin and lymphoproliferative disorders are two potential toxicities that are consistent with the immunosuppressive mechanism of action of tacrolimus, that have been reported in association with systemic tacrolimus use, and that were detected in preclinical studies of 0.03% and 0.1% tacrolimus ointment. In patients who receive systemic tacrolimus, it is hypothesized that chronic UV-light exposure is the principal co-factor for skin carcinogenesis, and that Epstein-Barr Virus (EBV) infection is the principal co-factor for lymphoproliferative disorders. It is important to note that these toxicities were **not** detected in the clinical trials for topical tacrolimus, but they are potentially life-threatening toxicities and are not necessarily reversible upon discontinuation of tacrolimus use. Sponsor has calculated that a human use safety margin from these toxicities should exist, based on a comparison of human bioavailability observed in the clinical trials with the bioavailability in animals in the toxicology studies (see Pharmacology/Toxicology Executive Summary). The existence of this safety margin is predicated on the assumption that the human carcinogenic risk is not augmented if tacrolimus is applied to dermatitic skin, and the validity of this assumption is unclear.

4. Considerations for the Advisory Committee

Given the uncertainty of the above potential risks to patients, particular consideration must be given to the necessity or utility of exercising the following risk management options: restricting the duration of use, restricting use to second-line therapy, mandating serologic testing for past or current EBV exposure, and encouraging sunlight avoidance. If the NDA is approved, there must also be considerable thought given to post-marketing studies needed to address the informational needs relating to the potential risks associated with this therapy.

APPENDIX 1

1- Primary Efficacy Results:

A- Success in the General Population:

Results for success at the end of treatment are summarized for the pediatric study in Table 1.

Table 1: Success Rate at the End of Treatment

Study	Treatment Group		
	Vehicle	Concentration of Tacrolimus Ointment	
		0.03%	0.1%
Intent-to-Treat Population[+]			
Pediatric Study	8/116 (6.9%)	42/117 (35.9%)	48/118 (40.7%)
Efficacy Evaluable Population[*]			
Pediatric Study	8/101 (7.9%)	39/108 (36.1%)	44/107 (41.1%)
Per Protocol Population[^]			
Pediatric Study	7/83 (8.4%)	35/91 (38.5%)	40/100 (40.0%)

[+]defined as all randomized patients who were dispensed study medication
[*]defined as all randomized patients who received study drug for at least 3 consecutive days (minimum 5 applications) beginning on Day 1 and had at least one "on treatment" value for the Physician's Global
[^]defined as all randomized patients who completed the study without a major protocol deviation as determined during a blinded patient classification review
Source: Table 5 of ISE and Table 8 of individual study report as amended in 11/9/99 and 4/21/00 submissions.

Results for P values of differences in success at the end of treatment are summarized in Table 2.

Table 2: Test of Significance for Success Rate

	P-Value†			
	Overall	0.03% vs Vehicle	0.1% vs Vehicle	0.03% vs 0.1%
Intent-to-Treat Population				
Pediatric Study	<0.001	<0.001	<0.001	0.401
Efficacy Evaluable Population				
Pediatric Study	<0.001	<0.001	<0.001	0.406
Per Protocol Population				
Pediatric Study	<0.001	<0.001	<0.001	0.755

Source: Table 6 of ISE and Table 9 of each individual study report as amended in 11/9/99 and 4/21/00 submissions.

Comments: For all three populations (MITT, efficacy evaluable, and per protocol population), a statistically significant difference in success was observed among the three treatment groups. A significantly greater success rate was observed for each tacrolimus ointment treatment group compared with the vehicle.

Table 6: Test of Significance for Success Rate: Gender

	P-Value from Cochran-Mantel-Haenszel Statistics			
	Overall	**0.03% vs Vehicle**	**0.1% vs Vehicle**	**0.03% vs 0.1%**
Males	<0.001	<0.001	<0.001	0.146
Females	<0.001	<0.001	<0.001	0.905

Table 7: Success Rate By Race (White, Black, Oriental)

	Treatment Group		
	Vehicle	**Concentration of Tacrolimus Ointment**	
		0.03%	**0.1%**
White	5/78 (6.4%)	30/76 (39.5%)	34/75 (45.3%)
Black	0/28 (0.0%)	8/32 (25.0%)	9/34 (26.5%)
Oriental	3/8 (37.5%)	3/7 (42.9%)	5/6 (83.3%)

Table 8: Test of Significance for Success: Race

	P-Value from Cochran-Mantel-Haenszel Statistics			
	Overall	**0.03% vs Vehicle**	**0.1% vs Vehicle**	**0.03% vs 0.1%**
White	<0.001	<0.001	<0.001	0.420
Black	0.014	0.006	0.004	0.861
Oriental	0.263	0.872	0.113	0.194

Table 9: Success Rate By Baseline Disease Severity

	Treatment Group		
	Vehicle	**Concentration of Tacrolimus Ointment**	
		0.03%	**0.1%**
Moderate	6/47 (12.8%)	20/45 (44.4%)	20/43 (46.5%)
Severe	2/69 (2.9%)	22/72 (30.6%)	28/75 (37.3%)

Table 10: Test of Significance for Success Rate: Baseline Disease Severity

	P-Value from Cochran-Mantel-Haenszel Statistics			
	Overall	**0.03% vs Vehicle**	**0.1% vs Vehicle**	**0.03% vs 0.1%**
Moderate	0.001	0.001	<0.001	0.727
Severe	<0.001	<0.001	<0.001	0.374

Table 11: Success By Percent BSA Affected at Baseline

	Treatment Group		
	Vehicle	Concentration of Tacrolimus Ointment	
		0.03%	0.1%
10-≤25%	4/33 (12.1%)	18/41 (43.9%)	11/27 (40.7%)
>25-≤50%	4/30 (13.3%)	11/27 (40.7%)	19/36 (52.8%)
>50-≤75%	0/25 (0.0%)	10/28 (35.7%)	14/34 (41.2%)
>75-100%	0/28 (0.0%)	3/21 (14.3%)	4/21 (19.0%)

Table 12: Test of Significance for Success Rate: Percent BSA Affected at Baseline

	P-Value from Cochran-Mantel-Haenszel Statistics			
	Overall	0.03% vs Vehicle	0.1% vs Vehicle	0.03% vs 0.1%
10-≤25%	0.009	0.003	0.024	>0.999
>25-≤50%	0.004	0.020	0.001	0.376
>50-≤75%	0.001	0.001	<0.001	0.598
>75-100%	0.065	0.030	0.021	0.685

Comments:

The results in Tables 3-12 demonstrate that:

1- The number of oriental subjects is very small (6 to 8 in each arm) resulting in failure to show effectiveness of either 0.03% or 0.1% formulations.

2- In all sub-populations, except for the oriental race, both the 0.03% and 0.1% formulations were significantly better than placebo.

3- The 0.1% formulation is not significantly better than the 0.03% formulation in any of sub-populations tested.

2- Secondary Efficacy Results:

The protocol-specified secondary efficacy variables included the change from baseline to the end of treatment for the Eczema Area and Severity Index (EASI), percentage of body surface area affected (% BSA), the Physician's Assessment of Individual Signs of Atopic Dermatitis, the Patient's Assessment of Treatment Effects (Overall Response and Pruritus), and the incidence of recurrence. Certain of these secondary efficacy variables may be relevant to the labeling.

A- Reduction in percentage of body surface area affected (% BSA)

The change from baseline to the end of treatment in the percentage of affected body surface area in the ITT population is presented in the following 2 tables.

Table 13: Change from Baseline to the End of Treatment: Affected Body Surface Area

Least Square Mean ± SE	Treatment Group		
	Vehicle	Concentration of Tacrolimus Ointment	
		0.03%	0.1%
Pediatric Study	N=116	N=117	N=118
Change from Baseline	-6.4 ± 1.98	-26.4 ± 1.90	-27.5 ± 1.91

Patient Population: Modified intent-to-treat; all randomized patients who received at least one dose of study drug (= all patients who were dispensed study drug).
Source: Section 8.3.6 (ISE Statistical Appendix 8.3.6.7.3).

Table 14: Test of Significance for Change from Baseline in Affected Body Surface Area

	P-Value from General Linear Model Analysis†			
	Overall	0.03% vs Vehicle	0.1% vs Vehicle	0.03% vs 0.1%
Pediatric Study	<0.001	<0.001	<0.001	0.664

Patient Population: Modified intent-to-treat; all randomized patients who received at least one dose of study drug (= all patients who were dispensed study drug).
† Statistical significance is indicated by p-values ≤0.05.
Source: Section 8.3.6 (ISE Statistical Appendix 8.3.6.7.3).

Comments: Statistically significantly greater improvement was observed for each tacrolimus ointment treatment group compared with the vehicle group. A statistically significant difference between tacrolimus ointment treatment groups was not observed in this study.

B- Reduction in physician assessment of individual signs

The Change from Baseline to the End of Treatment for Individual Signs of Atopic Dermatitis (ITT Population), and the results of the test of significance for Individual Signs (ITT Population), are shown in tables 15 and 16, respectively.

Table 15: Change from Baseline to the End of Treatment for Individual Signs of Atopic Dermatitis: Intent to Treat Population

Change from Baseline	Treatment Group		
	Vehicle	Concentration of Tacrolimus Ointment	
		0.03%	0.1%
Edema			
N	116	117	118
Least Squares Mean ± SE	-0.2 ± 0.06	-0.7 ± 0.06	-0.8 ± 0.06
Erythema			
N	116	117	118
Least Squares Mean ± SE	-0.2 ± 0.06	-0.8 ± 0.06	-0.8 ± 0.06
Excoriation			
N	116	117	118
Least Squares Mean ± SE	-0.2 ± 0.06	-0.7 ± 0.06	-0.9 ± 0.06
Lichenification			
N	116	117	118
Least Squares Mean ± SE	-0.2 ± 0.06	-0.8 ± 0.05	-0.7 ± 0.06
Oozing			
N	116	117	118
Least Squares Mean ± SE	-0.0 ± 0.05	-0.5 ± 0.05	-0.5 ± 0.05
Scaling			
N	116	1317	118
Least Squares Mean ± SE	-0.3 ± 0.06	-0.9 ± 0.06	-1.0 ± 0.06

Patient population: Intent-to-treat = modified intent-to-treat = all randomized patients who received at least one dose of study drug.
SE: standard error. Source: Integrated Summary of Effectiveness Appendix 8.3.6.7.3

Table 16: Test of Significance for Individual Signs of Atopic Dermatitis: Intent to Treat Population

Parameter	General Linear Model Analysis p-Values			
	Overall	0.03% vs Vehicle	0.1% vs Vehicle	0.03% vs 0.1%
Edema	<0.001	<0.001	<0.001	0.712
Erythema	<0.001	<0.001	<0.001	0.712
Excoriation	<0.001	<0.001	<0.001	0.152
Lichenification	<0.001	<0.001	<0.001	0.448
Oozing	<0.001	<0.001	<0.001	0.920
Scaling	<0.001	<0.001	<0.001	0.509

Patient population: Intent-to-treat = modified intent-to-treat = all randomized patients who received at least one dose of study drug. [One patient randomized to vehicle was never dispensed study drug; therefore modified intent-to-treat definition = FDA intent-to-treat definition].
Source: Integrated Summary of Effectiveness Appendix 8.3.6.7.3

Comments:

The representative score for each of the six clinical signs, edema, erythema, excoriation, lichenification, oozing, and scaling, was defined as the sum of the individual scores for all body regions treated at baseline divided by the number of regions treated at baseline. As shown in

tables 15 and 16, the results obtained for each of the six individual signs of atopic dermatitis showed significantly greater improvements in the 0.03% and the 0.1% tacrolimus ointment group compared to the vehicle. However, comparison of the 0.1% tacrolimus ointment group with the 0.03% tacrolimus ointment group failed to show any significant differences in the representative scores.

C- Patient's assessment of treatment effects

The treatment effects evaluated by the patients included the overall response and pruritus. Although both were included in the secondary efficacy criteria, the sponsor did not discuss the results of pruritus assessment by the patients in the ITT or MITT populations in the main text of the individual study reports or the ISE. Tables provided by the sponsor in the statistical appendices were used by the reviewer to evaluate this parameter in the ITT population (see comments).

The results of the patient's assessment of overall response were presented in the NDA, section 8.3.6 (ISE Statistical Appendix 8.3.6.6.3). These results were discussed in the NDA section 8.3.3.9 (ISE) and were summarized in table 25 of this section. Statistically significant differences were observed among treatment groups for the pediatric study ($p<0.001$ for each; CMH statistics testing for row mean score difference); statistically significant differences between each tacrolimus ointment group and vehicle were also observed ($p<0.001$ for each). No statistically significant differences were observed between 0.1% tacrolimus ointment and 0.03% tacrolimus.

Comments: The results of the patient's assessment of overall response are in general agreement with the primary efficacy variable.

The following table of the change in patient's assessment of pruritus has been compiled by the reviewer from the NDA statistical appendix 8.3.6.7.3 (ISE).

The differences shown in this table between the 0.03% or the 0.1% tacrolimus ointments and the vehicle were statistically significant, but the differences between the 0.03% and the 0.1% tacrolimus ointments were not statistically significant.

Table 17: Change from Baseline to the End of Treatment for Patient's Assessment of Pruritus: Intent to Treat Population

Change from Baseline	Treatment Group		
	Vehicle	Concentration of Tacrolimus Ointment	
		0.03%	0.1%
Protocols 97-0-037			
N	116	116	116
Least Squares Mean ± SE	-0.8 ± 0.30	-3.9 ± 0.29	-3.9 ± 0.29

Patient population: Intent-to-treat = modified intent-to-treat = all randomized patients who received at least one dose of study drug.
Source: Integrated Summary of Effectiveness Appendix 8.3.6.7.3.

APPENDIX 2

See data in the next two tables taken from the sponsor's report on the pivotal pediatric study, Appendix 14.2.2.5.1 and Table 14.3.5.2.1.

APPENDIX 14.2.2.5.1
KAPLAN-MEIER ESTIMATES OF TIME TO SLIGHT IMPROVEMENT
EFFICACY EVALUABLE POPULATION
CUMULATIVE IMPROVEMENT RATES BY VISIT

	TREATMENT GROUP		
VISIT	VEHICLE (N=101) RATE (SE)	0.03% (N=108) RATE (SE)	0.1% (N=107) RATE (SE)
WEEK 1 (UP TO DAY 11)	36.63(4.794)	74.07(4.217)	81.31(3.769)
WEEK 2 (UP TO DAY 18)	43.56(4.934)	84.26(3.504)	86.92(3.260)
WEEK 3 (UP TO DAY 25)	54.46(4.955)	89.81(2.910)	89.72(2.936)
WEEK 6 (UP TO DAY 50)	57.43(4.920)	91.67(2.659)	92.52(2.543)
WEEK 9 (UP TO DAY 71)	59.41(4.886)	93.52(2.369)	93.46(2.390)
WEEK 12 (UP TO DAY 92)	60.40(4.866)	93.52(2.369)	94.39(2.224)

IMPROVEMENT IS DEFINED BASED ON PHYSICIANS GLOBAL EVALUATION OF CLINICAL RESPONSE.
CUMULATIVE RATES ARE BASED ON KALPAN-MEIER ESTIMATES.

TABLE 14.3.5.2.1
TIME TO FIRST IMPROVEMENT OF PHYSICIANS GLOBAL EVALUATION
EFFICACY EVALUABLE POPULATION

Parameter	Class	TREATMENT GROUP VEHICLE (N=101)	0.03% (N=108)	0.1% (N=107)
>= SLIGHT IMPROVEMENT (DAYS)	N	61	101	101
	MEAN	16.4	11.6	10.9
	STD	15.4	9.8	10.5
	MIN	7	5	4
	Q1	8	8	8
	MEDIAN	9	8	8
	Q3	22	10	8
	MAX	85	69	78
>= MODERATE IMPROVEMENT (DAYS)	N	37	89	93
	MEAN	24.7	16.7	14.7
	STD	24.2	16.9	14.1
	MIN	8	7	4
	Q1	8	8	8
	MEDIAN	15	9	8
	Q3	25	18	15
	MAX	85	86	85
>= MARKED IMPROVEMENT (DAYS)	N	22	70	71
	MEAN	33.9	24.8	30.3
	STD	29.4	22.4	26.9
	MIN	8	7	7
	Q1	11	8	8
	MEDIAN	19.5	15	20
	Q3	46	28	43
	MAX	92	86	102
>= EXCELLENT IMPROVEMENT (DAYS)	N	11	44	46
	MEAN	26.7	40.5	47.2
	STD	24.8	26.3	30.8
	MIN	8	8	7
	Q1	11	15	15
	MEDIAN	15	43	56
	Q3	26	64	71
	MAX	85	94	102

APPENDIX 3

Table 9 Daily Hazard Rates* Over Time For Adverse Events - Long-Term Studies And Short-Term Studies** (MITT Population in Tacrolimus 0.1%)

COSTART TERM	DAY 1- 90 HAZARD(SE)	DAY 91-182 HAZARD(SE)	DAY 183-366 HAZARD(SE)
PROCEDURAL COMPLICATION	0.000 ( )	0.023 (0.0232)	0.044 (0.0312)
AORTIC STENOSIS	0.000 ( )	0.000 ( )	0.022 (0.0220)
GASTROINTESTINAL HEMORRHAGE	0.000 ( )	0.000 ( )	0.022 (0.0220)
RECTAL DISORDER	0.000 ( )	0.000 ( )	0.022 (0.0220)
LYMPHADENOPATHY	0.075 (0.0338)	0.093 (0.0466)	0.111 (0.0497)
HYPERCHOLESTEREMIA	0.000 ( )	0.000 ( )	0.022 (0.0220)
HYPOGLYCEMIA	0.000 ( )	0.000 ( )	0.044 (0.0311)
HYPOMAGNESEMIA	0.000 ( )	0.000 ( )	0.022 (0.0220)
DEPRESSION	0.015 (0.0151)	0.046 (0.0328)	0.044 (0.0312)
HYPERTONIA	0.000 ( )	0.023 (0.0231)	0.022 (0.0220)
HYPOTONIA	0.000 ( )	0.000 ( )	0.022 (0.0220)
SLEEP DISORDER	0.000 ( )	0.000 ( )	0.022 (0.0220)
THINKING ABNORMAL	0.000 ( )	0.000 ( )	0.022 (0.0220)
SEBORRHEA	0.000 ( )	0.023 (0.0232)	0.066 (0.0381)
KERATITIS	0.000 ( )	0.023 (0.0232)	0.044 (0.0312)

* Hazard Rate (x1000) For 1-90 Day, 91-182 Day And 183-366 Day Based On The Life Table Method.
** Long-Term Studies: Fg06-12 And 96-0-025, Short-Term Studies: 97-0-035, 97-0-036 And 97-0-037.
Source: Attachment 3 of Vol.1 of NDA 50777 submitted on 4/21/00.

PHARMACOLOGY/TOXICOLOGY
TAB 2

EXECUTIVE SUMMARY
NDA 50-777; Protopic (Tacrolimus) Ointment; Atopic Dermatitis
Pharmacology/Toxicology Summary

INTRODUCTION AND DRUG HISTORY:

Tacrolimus (also known as FK506) is a 23 member macrolide immunosuppressant produced by *Streptomyces tsukubaensis*, a soil bacterium found in Mount Tsukba, Japan. Tacrolimus inhibits the early activation of T-lymphocytes. Tacrolimus was originally approved in the United States in April 1994 in Prograf® capsules (NDA 50-708) and injection (NDA 50-709) for the prophylaxis of organ rejection in patients receiving allogenic liver transplants. Supplemental NDAs 50-708 (S-008)/50-709 (S-009) were approved in April 1997 for prophylaxis of rejection after allogenic kidney transplantation.

A topical formulation of tacrolimus, Protopic® (tacrolimus) ointment, has been developed by Fujisawa for dermatologic use. Protopic® ointment is indicated for the treatment of atopic dermatitis. The rationale for this is that atopic dermatitis is considered an immunologic disorder believed to be modified by T-lymphocytes. The sponsor plans to market two strengths of the Protopic® ointment (0.03% and 0.1%).

The nonclinical pharmacology/toxicology of orally or intravenously administered tacrolimus has been established under NDAs 50-708/50-709. Potential target organs of toxicity identified in these studies included kidneys, pancreas, thymus, lymph nodes and spleen. The sponsor submitted IND the division in December 1994 for studying the efficacy and safety of Protopic® ointment in the treatment of atopic dermatitis. Additional nonclinical pharmacology/toxicology studies were conducted with tacrolimus ointment under IND to support the safety of topical application of Protopic® ointment.

CHRONIC TOXICOLOGY:

Repeat dose toxicology studies for topically applied tacrolimus ointment were conducted for a duration up to 26 weeks in rats and 52 weeks in micropigs under NDA 50-777. Topical treatment of rats with tacrolimus ointment for 26 weeks generated similar results noted after oral administration in rats. Target organs of toxicity identified in this study included kidneys, thymus, spleen, pancreas, cervical lymph nodes and bone marrow. Treatment related mortalities were noted in the 0.5% and 0.3% tacrolimus ointment treatment groups. The systemic toxicity effects noted in this study were due to the substantial level of cutaneous absorption noted through rat skin. Tacrolimus blood concentrations at 6 hours post application of the 0.5% ointment (10 mg/kg/day) were 4.2 ng/ml and 5.0 ng/ml at week 13 and week 26, respectively. For comparison purposes, the mean tacrolimus blood concentration for a 10 mg/kg/day dose in a 13 week oral (dietary admix) toxicity study in mice was 7 - 8 ng/ml. No treatment related dermal reactions were observed macroscopically at the treated skin sites in this study. Microscopically noted dermal reactions at the treatment site included a non-dose related incidence of epithelial hyperplasia/acanthosis. The no observed adverse effect dose (NOAEL) in rats administered topical tacrolimus ointment daily for 26 weeks was identified as 0.03% (0.6 mg/kg/day; 1.8

mg/m^2/day) in this study based on the systemic toxicity profile. The NOAEL dose is 25X the maximum recommended human dose (MRHD; 44.4 mg/m^2/day) based on body surface area comparisons.

Topical treatment of micropigs with tacrolimus ointment for 52 weeks generated much less toxicity than was noted in rats. The reason for this is probably related to significantly less systemic absorption through the skin of the minipig compared to the rat. The level of systemic absorption after topical application of tacrolimus ointment in micropigs more approximates that noted in humans. Treatment related dermal effects were noted in this study that exhibited a similar incidence and severity for vehicle ointment treated and tacrolimus ointment treated animals. The macroscopic changes included papules, circular purple ring, hyperpigmentation and hypopigmentation. Most of these changes corresponded microscopically to epidermal acanthosis with hyperkeratosis and perivascular mononuclear cell infiltrates in the papillary dermis consistent with hyperplastic dermatitis. The histopathological findings in the treatment site skin were noted with about the same incidence in vehicle ointment and tacrolimus ointment treated animals. Therefore, the dermal effects noted in this study are probably related to vehicle ointment rather that tacrolimus. Several of the microscopic alterations in the vehicle ointment treated and tacrolimus ointment treated animals were characteristic of those described for human paraffin (oil) dermatitis. These findings included hyperkeratosis, hyperpigmentation, epidermal pustules, chronic perivascular inflammation and follicular changes (dilatation and hyperkeratosis at the infundibulum). The ointment base contains propylene carbonate, white beeswax, 11% liquid paraffin (mineral oil), hard paraffin, and white petrolatum. Therefore, a possible cause of the skin lesions in this study may be due to the presence of liquid paraffin in the ointment base. The only systemic toxicity noted in this study was a significantly lower body weight in females only (noted after week 19) treated with 3.0% tacrolimus ointment. The NOAEL in male micropigs administered topical tacrolimus ointment twice daily for 52 weeks was 3.0% (18 mg/kg/day; 486 mg/m^2/day; Month 6 $AUC_{0\text{-}24\ hr}$ = 185 ng·hr/ml) and for female micropigs the NOAEL was 1.0% (6 mg/kg/day; 162 mg/m^2/day; Month 6 $AUC_{0\text{-}24\ hr}$ = 168 ng·hr/ml) in this study. The NOAEL dose is 8-9X the MRHD based on AUC levels (human $AUC_{0\text{-}24\ hr}$ = 20.4 ng·hr/ml for 0.1% tacrolimus ointment; based on European clinical study data discussed in more detail in the clinical relevance of safety issues section below).

IMPURITY TOXICOLOGY:

The sponsor identified an impurity in the tacrolimus ointment as and stated that the level of this impurity was The impurity has been characterized as one of the tacrolimus derivatives in which the structure was exchanged from The sponsor has conducted five nonclinical toxicology studies for qualification of the impurity under NDA 50-777. The sponsor determined that expressed similar acute intravenous toxicity as tacrolimus in rats, was a non-irritant at 0.3% concentration in rabbits and was not mutagenic in the Ames test or clastogenic in the chromosomal aberration assay. The sponsor also conducted a 4 week repeat dermal toxicity study in rats comparing the toxicity profile of ointment and 1% tacrolimus ointment to vehicle ointment and sham control animals, per the division's request. In this study the 1% tacrolimus ointment exhibited a toxicity profile consistent with what has been observed in previous studies. However, no toxicity was noted in th ointment treated animals in this study. This result suggests that the impurity

does not contribute to the toxicity profile of the tacrolimus ointment. The results from the 5 toxicity studies conducted with the impurity serve to qualify the impurity in the tacrolimus ointment.

REPRODUCTIVE TOXICOLOGY:

The reproductive toxicity of orally administered tacrolimus was evaluated in Segment 1 (rats), Segment 2 (rats and rabbits) and Segment 3 (rats) studies in NDAs 50-708/50-709. Orally (gavage) administered tacrolimus altered reproductive function in female animals and reduced offspring viability during reproductive toxicity studies with rats and rabbits. Male reproductive behavior was slightly altered in rats and rabbits. The changes in reproductive parameters observed during these studies included increased copulatory intervals, decreased implantation, increased loss of fetuses, fewer births, and smaller litter sizes. No reduction in male or female fertility was evident. Adverse effects in offspring whose mothers received tacrolimus during pregnancy included markedly reduced viability and slightly increased incidence of malformation. Based on the results of these studies, oral tacrolimus was labeled as pregnancy category C. The sponsor did not conduct any nonclinical topical application reproductive toxicology studies under NDA 50-777. Therefore, the pregnancy category will remain C and the information contained in the appropriate section of the label that addresses reproductive toxicity that was included in the Prograf®(oral tacrolimus) label will be incorporated into the Protopic® (topical tacrolimus) label.

GENOTOXICITY:

Under NDAs 50-708/50-709 tacrolimus had undergone testing in a battery of genotoxicity tests. No evidence of genotoxicity was seen in bacterial (Salmonella and E. Coli) or mammalian (Chinese hamster lung-derived cells) *in vitro* assays of mutagenicity, the *in vitro* CHO/HGPRT assay of mutagenicity, or *in vivo* clastogenicity assays performed in mice. Tacrolimus did not cause unscheduled DNA synthesis in rodent hepatocytes. In summary, tacrolimus showed no evidence of genotoxic potential.

CARCINOGENICITY:

Two oral (feed) carcinogenicity studies in mice and rats have been conducted for tacrolimus under NDA 50-708/50-709. The results from these two studies were negative but there is some question as to whether the systemic exposure was adequate in these two studies. The sponsor was requested by the division to conduct a 2 year dermal carcinogenicity study in mice and a one year photocarcinogenicity study in hairless mice to support the tacrolimus ointment formulation.

The results of the one year photocarcinogenicity study conducted in hairless mice demonstrated that topical administration of the vehicle with UV radiation exposure greatly enhanced photocarcinogenesis (defined as time in weeks to median tumor onset of skin papillomas ≥ 1 mm). The vehicle induced enhancement tended to be greater in male mice as compared to female mice. Topical administration of tacrolimus ointment (0.03%, 0.1%, 0.3% and 1.0%) had an additional small influence on tumor development beyond the vehicle effect,

which was more prevalent in male mice. A dose dependent increase in mortality unrelated to tumor burden was noted after topical administration of the tacrolimus ointment. Significant systemic absorption was obtained after dermal application of tacrolimus ointment to hairless mice. The high level of systemic absorption is confirmation that the signs of systemic toxicity noted in this study are due to relatively high systemic exposure to tacrolimus.

The data for the time to median tumor onset for tumors ≥1 mm from the nonclinical photocarcinogenicity study is provided in the following table.

Tacrolimus (%)	UVR Exposure (RBU/Week)	Male Mice Median (Weeks)	Female Mice Median (Weeks)
None (Untreated)	600	44.5	42
0 (Vehicle)	600	33.5[c]	36[a]
0.03	600	31.5[c]	44
0.1	600	30[c,1]	42
0.3	600	30[c,2]	34[a]
1.0	600	30.5[c,2]	32[b]
None (Untreated)	1020	28[c,3]	28.5[c,3]

a = $p<0.05$ compared to untreated control; b = $p<0.01$ compared to untreated control; c = $p<0.001$ compared to untreated control.
1 = $p<0.05$ compared to vehicle control; 2 = $p<0.01$ compared to vehicle control; 3 = $p<0.001$ compared to vehicle control.

In the two year dermal mouse carcinogenicity study, high levels of mortality were exhibited in the 0.3%, 1.0% and 3.0% tacrolimus ointment dose groups. All animals died by week 26 in the 3.0% dose group and by week 46 in the 1.0% dose group. Approximately 85% of animals died by the end of the study in the 0.3% dose group. Adequate numbers for statistical evaluation of tumor incidence were available in the sham control, vehicle control, 0.03% and 0.1% tacrolimus ointment groups. The maximum tolerated dose (MTD) was identified as the 0.1% tacrolimus ointment dose based on mortality. The neoplastic findings noted in this study are provided in the following table.

Neoplastic Findings in Tacrolimus Ointment Treated Mice
(Including Animals Found Dead)

Note: Neoplastic incidences from the scheduled sacrifice are listed as the first numbers for each tumor type. Neoplastic incidences from unscheduled sacrifices or animals found dead are listed as the second numbers for each tumor type. Total values for scheduled and unscheduled tumor incidences are in parentheses as the third number for each tumor type.

	Males				Females			
	Untreated	Vehicle	0.03%	0.1%	Untreated	Vehicle	0.03%	0.1%
Hemolymphoretic System								
Lymphoma, Pleomorphic (Total)	6/42 1/8 **(7/50)**	2/41 0/9 **(2/50)**	3/39 1/11 **(4/50)**	14/21 11/29 **(25/50)**	11/41 1/9 **(12/50)**	5/44 1/6 **(6/50)**	12/39 2/11 **(14/50)**	17/20 10/30 **(27/50)**
Lymphoma, Undifferentiated (Total)	0/42 0/8 **(0/50)**	1/41 0/9 **(1/50)**	0/39 2/11 **(2/50)**	0/21 4/29 **(4/50)**	2/41 1/9 **(3/50)**	1/44 0/6 **(1/50)**	1/39 2/11 **(3/50)**	1/20 12/30 **(13/50)**

A statistically significant elevation in the incidence of pleomorphic lymphoma in high dose male (25/50; $p<0.0001$) and female animals (27/50; $p<0.0001$) and in the incidence of undifferentiated lymphoma in high dose female animals (13/50; $p=0.0005$) compared to vehicle control was noted in the mouse dermal carcinogenicity study.

Appropriate information from both the photocarcinogenicity and dermal carcinogenicity studies conducted with tacrolimus ointment is recommended for incorporation into the label.

CLINICAL RELEVANCE OF SAFETY ISSUES IDENTIFIED IN NONCLINICAL TOXICOLOGY STUDIES:

The major clinically relevant safety issues identified from the nonclinical toxicology studies conducted with tacrolimus ointment relate to the results of the photocarcinogenicity and dermal carcinogenicity studies. The decrease in time to skin tumor development noted in the mouse photocarcinogenicity study is a strong signal that tacrolimus ointment can potentially increase the risk of skin cancer from UV exposure in humans. Therefore, it is recommended that the label contain a warning to assure that patients minimize sunlight exposure during the use of tacrolimus ointment.

The significant elevation in the incidence in lymphoma noted in the mouse dermal carcinogenicity study is potentially cause for significant concern. One component involved in the final decision on whether this is a significant safety concern for use of tacrolimus ointment in humans is dependent on the extent of systemic exposure that occurs at the maximum recommended human dose. The highest proposed dose for tacrolimus ointment is the 0.1% concentration. This is the concentration level that was the MTD in the dermal carcinogenicity study and the concentration at which a statistical increased incidence of pleomorphic and undifferentiated lymphoma was noted in mice. Significant systemic absorption of tacrolimus occurs in mice after topical administration of tacrolimus ointment. Human pharmacokinetic data available from the original NDA submission for tacrolimus ointment provided AUC data for the 0.3% tacrolimus ointment. Only whole blood levels were available from the 0.03% and 0.1% tacrolimus ointment concentrations.

During a team review meeting conducted on 3-29-00 to discuss the results of the mouse dermal carcinogenicity study, one of the issues discussed was the need for human AUC data for the 0.03% and 0.1% tacrolimus ointment concentrations. The clinical pharmacology reviewer

recommended that the sponsor conduct another human pharmacokinetic study to obtain this data directly. In addition, the sponsor may be able to provide a rough estimate of the AUC data from the collected plasma levels obtained in the clinical studies conducted to date with tacrolimus ointment. It was recommended that the sponsor perform such a calculation and submit the AUC data to the division for review. This rough estimate would be useful in providing an estimate of the safety factor for lymphoma formation after topical application of tacrolimus ointment in humans.

The sponsor provided data (in a submission to NDA 50-777 dated 6-5-00) from a European clinical pharmacokinetic study in which atopic dermatitis patients were treated with 0.1% tacrolimus ointment. The highest mean $AUC_{0\text{-}12\ hr}$ value observed in the adult study was 10.2 ng·hr/ml on day 4 in the group with 36-60% of body surface area treatment (n=9). This would equal an $AUC_{0\text{-}24\ hr}$ value of 20.4 ng·hr/ml. The no effect dose AUC in the dermal carcinogenicity study is ~9 fold greater that the maximum human AUC obtained in this study (189 ng·hr/ml ÷ 20.4 ng·hr/ml). The AUC for the dose that lymphomas were noted in the dermal carcinogenicity study is ~26 fold greater than the maximum human AUC obtained in this study (534 ng·hr/ml ÷ 20.4 ng·hr/ml).

Human patients may not have a high risk of getting lymphomas under conditions of clinical use for the 0.03% and 0.1% tacrolimus ointment based on the fold level calculations. However, human patients may have a higher risk of developing skin cancer with the use of 0.03% and 0.1% tacrolimus ointment based on the results of the nonclinical photocarcinogenicity assay. Therefore, it is recommended that a warning be included in the label for patients to minimize or avoid exposure to natural or artificial sunlight during the use of 0.03% and 0.1% tacrolimus ointment.

CLINICAL PHARMACOLOGY / BIOPHARMACEUTICS

TAB 3

Executive Summary of Clinical Pharmacokinetics Review

I. BACKGROUND

Dosage Form: Ointment for topical application (0.03% and 0.1%)
(Note: 0.3% ointment was used in the initial dose development stages but was not used as the final to-be marketed strength in the U.S.)

Indication: For the treatment of atopic dermatitis in adults and pediatric patients 2 years of age and older.

Pharmacologic Class: A macrolide immunosuppresant produced by *Streptomyces tsukubaensis* (a soil bacterium found in Mount Tsukuba, Japan). It is also known as FK506. Tacrolimus inhibits the early activation of T-lymphocytes resulting in decreased production of cytokines. Atopic dermatitis is considered to be an immunologic disorder believed to be modified by T-lymphocyte activation.

Clinical Endpoints: Primary efficacy end point was incidence of success, defined as rating of cleared or excellent improvement (≥90% improvement of areas defined for treatment at baseline based on Physician's Global evaluation of Clinical Response). Secondary endpoints were eczema area and severity score, % body surface area affected, Physician's assessment of Individual Signs of Atopic Dermatitis and Patient's assessment of Pruritis.

Dosage and administration: Applied topically twice daily as a thin layer to affected areas of the skin. The use of occlusive dressings is not recommended.

Foreign marketing history: World-wide in both intravenous (Prograf®, NDA 50-708, April 1997) and oral formulations (Prograf®, NDA 50-709, April 1994) for the prevention of organ rejection following allogenic liver or kidney transplantation. 0.1% tacrolimus ointment has been approved for marketing in Japan since June 1999.

Formulation: Tacrolimus Ointment is an oleaginous ointment in which the drug substance is dissolved in droplets of propylene carbonate, which are uniformly dispersed in vehicle. The ointment will be supplied in 30 and 60 gram tubes. The composition of 0.03% and 0.1% is provided in the following table.

Ingredient	0.03% (%w/w)	0.1% (%w/w)
Tacrolimus	0.03	0.1
White Petrolatum, USP		
Mineral Oil, USP		
Propylene Carbonate, NF		
White Wax, NF		
Paraffin, NF		
Total	100	100

II. ANALYTICAL VALIDATION

Blood to plasma partitioning of tacrolimus is saturable and variable, therefore the pharmacokinetics of tacrolimus has been expressed in terms of blood concentration. The sponsor has analyzed tacrolimus in blood by two different methodologies with the lower limit of detection of tacrolimus given as below:

(i)
(ii)

III. PHARMACOKINETIC STUDIES

The sponsor has conducted a total of 5 pharmacokinetic studies in healthy volunteers or adult and pediatric patients with atopic dermatitis as highlighted below, with a brief description of the study design:

In healthy Volunteers:

(i) Study FG-06-04: A pharmacokinetic study single and multiple topical doses of 0.03, 0.1 and 0.3% tacrolimus ointment
Dose and duration: q.d. for 14 days
Area of application: 1000 cm^2 area of back
Total tacrolimus exposure during treatment period: 21-210 mg (15mg x14)
N= 12 (6M & 6F)

In adult and pediatric patients with atopic dermatitis

(ii) Study 94-0-008: A pharmacokinetic study in adult and pediatric patients following single and repeat application of 0.3% tacrolimus ointment
Dose and duration: q.d on Day 1& 8, b.i.d. on Days 2-7
Area of application: 100-5000 cm^2 area of trunks/limbs or face for adults
50-100 cm^2 of trunks/limbs in pediatrics
Total tacrolimus exposure during treatment period: 21, 105, 210 and 630 mg in adults and 10.5 and 21 mg in pediatrics
N= 31 adults (16M & 15F), 8 pediatrics (ages 5-11)

(iii) Study FJ-106: A study in adult patients with topical application of 0.1 and 0.3% tacrolimus ointment

Dose and duration: single dose or b.i.d. for 7 days
Area of application: not provided
Total tacrolimus exposure during treatment period: 1.25-10 mg single dose, 70 & 140 mg total after multiple doses
N= 21

(iv) Study FG-506-06-22: Pharmacokinetic study (dose escalation) in adult patients after single and multiple doses of 0.1% topical tacrolimus ointment
Dose and duration: b.i.d. for 13 days, single dose on Day 14
Area of application: ≤3000 cm^2, >3000≤6000 cm^2, >6000≤10000 cm^2 area
Total tacrolimus exposure during treatment period: 28.27-269.73 mg
N= 32 (8M & 24F)

(v) Study FG-506-06-23: Pharmacokinetic study in pediatric patients after single and multiple doses of 0.1% topical tacrolimus ointment (interim report)
Dose and duration: b.i.d. for 13 days, single dose on Day 14
Area of application: ≤1500 cm^2, >1500≤3000 cm^2, >3000≤5000 cm^2 area
Total tacrolimus exposure during treatment period: 5.4-154.44 mg
N= 20 (12M & 8F)

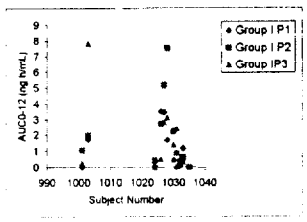
The following questions can be raised regarding the pharmacokinetics of topical 0.03% and 0.1% tacrolimus ointment. The questions raised are followed by the answers and important observations given as bullets as obtained from the review of the application.

Is tacrolimus systemically absorbed after topical application of 0.03% and 0.1% tacrolimus ointment? If yes, what is the exposure levels in adult and pediatric patients with atopic dermatitis?

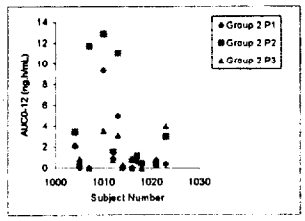
- Yes, tacrolimus is systemically absorbed after the topical application of 0.03% and 0.1% tacrolimus ointment in adult and pediatric patients with atopic dermatitis, although the systemic absorption is highly variable.

Area under the curve (AUC_{0-24} or AUC_{0-12}):

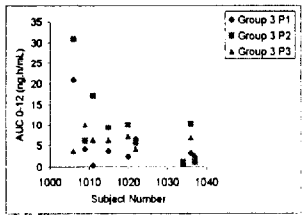
- The systemic exposure was low and highly variable in adult patients with AUC_{0-12} ranging between 0-30.97 ng.hr/mL or an AUC_{0-24} ranging between 0-61.94 ng.hr/mL and a highest mean AUC_{0-12} of 10.2 ng.h/mL after the twice daily topical application of 0.1% tacrolimus ointment for 13 days.
The AUC_{0-12} from Study FG-506-06-22 in three adult groups by increasing exposure is shown in the following figures, P1, P2 and P3 represent AUCs at Day 1, 4 and 14.



16±5% total BSA treated



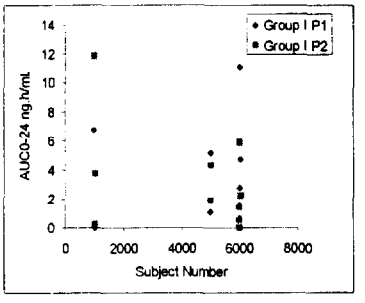
25±6% total BSA treated



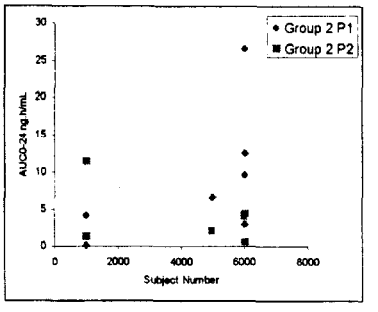
53±7% total BSA treated

The AUC_{0-24} after once daily topical application of 0.3% tacrolimus ointment (3 times more than the highest to-be marketed strength) ranged from 0.9 to 42.5 ng.hr/mL in adult patients.

- The systemic exposure was highly variable in pediatric patients (ages 5-12 years) with AUC_{0-24} ranging between 0-27 ng.h/mL and a highest mean AUC_{0-24} of 10.6 ng.h/mL, as shown in the figure below.



33±9% total BSA treated

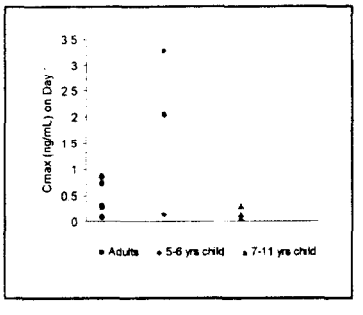


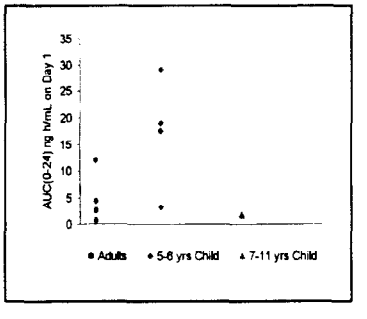
63±11% total BSA treated

Maximum blood level (C_{max}):

- The C_{max} was below 5 ng/mL in most adult pateints, with the exception of blood levels in three subjects of 5.5 ng/mL (on Day 14), 9.8 (on Day 4) and 15 ng/mL (on Day 14) after the topical application of 0.1% tacrolimus ointment. From Study FJ-106, the highest C_{max} obtained in two subjects was 20 ng/mL after single and multiple dose of 0.1% tacrolimus ointment. The C_{max} observed in adult patients from clinical trials was 14 ng/mL (from study FJ-111). The C_{max} observed in adult patients after the topical application of 0.3% tacrolimus ointment was 9.42 ng/mL.
- The C_{max} was below 1.6 ng/mL in all pediatric patients after the topical application of 0.1% tacrolimus ointment in a pharmacokinetic study. The highest C_{max} observed after the topical application of 0.3% tacrolimus ointment was 3.28 ng/mL. The highest C_{max} observed in the pediatric patients from the clinical trials was 9.58 ng/mL (Study 95-0-009).

  In a pharmacokinetic study with 0.3% tacrolimus ointment in pediatric patients it was observed that the younger children (5-6 years, N=4) had higher blood levels of tacrolimus as compared to older children (7-11 years, N=4) and adults, when treated on the same body parts (trunk and limbs), as shown in the figures below. The younger children were exposed to half the dose of the adults, but their C_{max} and AUCs were 2-3 times higher than that of the adults and older children.





No clinical significance can be derived on this small sample size of 4 pediatric patients in the age group 5-6 years and 7-11 years.
No obvious differences were seen when the samples from the clinical trials for evaluated closely for the tacrolimus blood levels in the younger children (ages 2-6 years). A pharmacokinetic study in children between the ages 2-4 has not been conducted.

- The sponsor has not studied the 0.03% strength of the tacrolimus ointment in a pharmacokinetic study, but this is acceptable because the 0.03% strength is lower than the 0.1% strength, which has been studied in a detailed study. The concentrations were below 5 ng/mL in most patients after the topical adminsitration of 0.1% ointment. Therefore, the blood levels of tacrolimus are very likely to be much lower after the topical application of the 0.03% ointment. However, the blood levels were evaluated in the Phase III clinical study with the highest observed concentration being 1.19 ng/ml in the pediatric population (Study #97-0-037) and 8.13 ng/mL in the adult patients (Study #97-0-036).

Accumulation:

- There was no systemic accumulation of tacrolimus following repeat application of 0.1% tacrolimus ointment for 2 weeks in adult or pediatric atopic dermatitis patients. However, after topical application of 0.3% tacrolimus ointment older children (7-11 years) had twice the C_{max} and AUC_{0-24} on Day 8 as compared to Day 1.

Systemic exposure under occlusion:

- The pharmacokinetic studies have not been conducted under occlusion.

Is the systemic absorption of tacrolimus similar in patients with atopic dermatitis and healthy volunteers?

- The systemic absorption is more (10 times) in patients as compared to healthy volunteers. The highest C_{max} observed in one healthy subject was 0.128 ng/mL after topical application of 0.1% tacrolimus ointment. The maximum concentration observed in a healthy volunteers applying 0.3% tacrolimus ointment was 0.127 ng/mL, where as the maximum concentration seen in patients receiving the same dose was 1.2 ng/mL (5 g of 0.3% in 1000 cm^2 area of exposure)

Are the systemic levels of tacrolimus similar in adult and pediatric patients with atopic dermatitis?

- There are no significant differences in systemic exposure between adult and pediatric patients (6-12 years of age) based on studies FG-506-22 and FG-506-23. The highest AUC_{0-24} observed in a 5 year old child from Study 94-0-008 was 29.2 ng.h/mL. No

obvious differences were seen when blood samples from Phase III clinical trials were evaluated closely for blood levels in younger children (ages 2-6 years). The highest C_{max} observed from clinical trials was 9.58 ng/ml from Study 95-0-009 in pediatric patients. The pharmacokinetic study (94-0-008) did show a trend towards higher concentration in the younger children (ages 5-6 years), but there were only 4 children in this study and a 3-10 times higher strength (0.3%) than a to-be-marketed strength (0.03 and 0.1%) was used in this study.

Is the systemic absorption of topical tacrolimus treatment site dependent?

- Tacrolimus ointment was more permeable through atopic dermatitis lesions on the face and neck regions as compared to the trunk and limb regions in a study with 0.3% tacrolimus ointment. The effect of treatment site has not been evaluated in children, only the trunks and limbs were evaluated in this study.
- Systemic exposure was the same on Day 1 and Day 8, when tacrolimus ointment was applied on the facial lesions, where as tacrolimus concentrations were lower on Day 8 when applied to the trunks and limbs in a 8 day study with 0.3% tacrolimus ointment.
- In the study with 0.1% tacrolimus ointment in adults and pediatric patients effect of treatment site has not been evaluated, the ointment was applied to the head & neck/trunks/upper limbs/lower limbs.

How does systemic absorption of tacrolimus compare after oral and topical administration of tacrolimus?

- The oral absolute bioavailability of tacrolimus is 15-25% (highly variable) and the absolute bioavailability of topical tacrolimus based on historical IV data is 0.5%.
- The comparative AUCs and C_{max} of tacrolimus in various types of adult and pediatric patients are shown in the following table. The oral data in the following table has been taken from the PDR® on Prograf®.

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Population	N	Route of Tacrolimus administration (Dose)	Highest mean AUC_{0-24} ng.h/mL	Highest Observed C_{max} ng/mL
Healthy	1	Topical (0.3% qd for 14 days)	2.31	0.127
Atopic Dermatitis Adult Patients	9	Topical (0.1% b.i.d for 13 days, 16-53% of total BSA[d])	20.4 ± 18.4	14.975 [a]
Atopic Dermatitis Pediatric Patients	20	Topical (0.1% b.i.d for 13 days, 33-61% of total BSA[d])	10.6± 15.0	1.513
Liver Transplant Pediatric Patients	9	Oral (0.15-0.2 mg/kg/day)	337±167	43.4±27.9[c]
Kidney Transplant Adult Patients	26	Oral (0.2 mg/kg/day)	486±172 [b]	41.2±17.3[c]
Liver Transplant Adult Patients	17	Oral (0.3 mg/kg/day)	519±179 [b]	-

[a]The highest concentration of 20 ng/mL was observed in 2 patients after single and multiple application of 0.1% tacrolimus ointment from another study. AUC calculations could not be done for this study.
[b]AUC_{0-inf}
[c]mean Cmax
[d]Body Surface Area

- Based on the evaluation of the data it is apparent that the systemic absorption of tacrolimus after topical application of 0.1% tacrolimus ointment is very low relative to the exposure generated from oral dosing. What is left unanswered is the amount of tacrolimus that enters the lymphatic system following topical administration. Drainage of the drug from the skin to the lymph system will be more relevant rather than the skin/blood partitioning.

What is known about the metabolism of tacrolimus after topical application?

- The metabolites of tacrolimus were below the limit of detection in healthy volunteers. The sponsor has not evaluated the presence of metabolite in patients. However, in the clinical study FG-06-19 in pediatrics, the levels of metabolites have been evaluated and submitted as an update of 120-day safety update. Trace levels of metabolite M-I (13-demethyl tacrolimus) were present, with no evidence of M-II (31-demethyl tacrolimus) in the samples. With the lower concentrations of tacrolimus observed upon topical administration, as compared to oral tacrolimus, this should not be of concern. In vitro studies indicated that tacrolimus is not metabolized in human skin.

CHEMISTRY TAB 4

Specification: Related Substances	Identified single peaks:	Range
	Others:	
	Unidentified single peaks:	
	Total:	

Impurity is an impurity seen only in the ointment, and not in the solid state or in solution. The applicant postulates that this impurity arises from of tacrolimus, which is known to have greater in solution. This structure has been proposed for this (the relative and absolute configurations were not specified):

Other impurities listed in the literature references were identified during characterization of the active ingredient for NDAs 50-708 and 50-709, and were previously qualified.

Clinical Formulations

Six concentrations of tacrolimus ointment were studied in preclinical and clinical trials: 0.03%, 0.1%, 0.3%, 0.5%, 1.0% and 3.0% (w/w), as well as a placebo formulation. The three higher concentrations were eliminated based on preclinical safety results, and the 0.3% dosage was used for dose development in Phase 2 trials, and subsequently dropped.

Based on the 0.1% strength and the 60g package size, the maximum exposure to tacrolimus is expected to be 0.06g (60mg) per tube.

The vehicles for the two concentrations proposed for marketing are almost identical: mineral oil, paraffin, propylene carbonate, white petrolatum, and white wax.

In Vitro release Testing

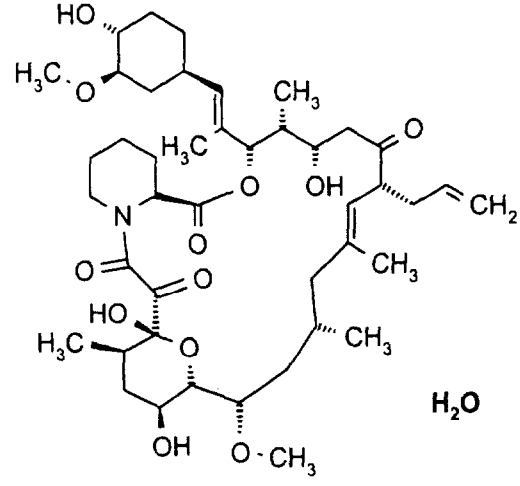
In vitro release tests and in vivo absorption studies were performed to evaluate potential differences in ointment products manufactured with

PROTOPIC® (tacrolimus) Ointment, 0.03% and 0.1% (w/w)
Chemistry, Manufacturing and Controls Executive Summary

PROTOPIC® (tacrolimus ointment) Ointment contains tacrolimus, a macrolide immunosuppressant produced by *Streptomyces tsukubaensis*. Each gram of PROTOPIC® Ointment contains either 0.03% or 0.1% (w/w) of tacrolimus (on dry basis) in a base of mineral oil, paraffin, propylene carbonate, white petrolatum and white wax. The drug product is packaged in 30g and 60g laminated tubes, and in a 3g physician's sample. The proposed expiration period for this drug product is 24 months.

Drug Substance

The active ingredient, tacrolimus, has the following structural formula and chemical name:



[3S-[3R*[E(1S*,3S*,4S*)],4S*,5R*,8S*,9E,12R*,14R*,15S*,16R*,18S*, 19S*,26aR*]]-5,6,8,11,12,13,14,15,16,17,18,19,24,25,26,26a-hexadecahydro-5,19-dihydroxy-3-[2-(4-hydroxy-3-methoxycyclohexyl)-1-methylethenyl]-14,16-dimethoxy-4,10,12,18-tetramethyl-8-(2-propenyl)-15,19-epoxy-3H-pyrido[2,1-c][1,4]oxaazacyclotricosine-1,7,20,21(4H,23H)-tetrone, monohydrate.

Tacrolimus has an empirical formula of $C_{44}H_{69}NO_{12} \bullet H_2O$ and a formula weight of 822.05.

This active ingredient is approved for PROGRAF® Capsules and for Injection (NDAs 50-708 and 50-709). Tacrolimus is manufactured by fermentation of *Streptomyces tsukubaensis*, and is purified by Tacrolimus is known to interconvert in solution to other isomers / tautomers, and the impurity profile has been extensively studied. A number of related substances and degradation products have been identified and reported in the chemistry literature (Namiki, Y., et al., Chromatographia, Vol. 40, No. 5/6, March, 1995), and these impurities are controlled by specifications for Related Substances:

different The in vitro release testing was performed

Ointments of 0.03%, 0.1% and 0.3% strengths were evaluated.

The release profile of the various strengths (release rate vs. concentration) was linear with respect to strength, indicating no differences in the diffusion coeffecient of tacrolimus in the different dosage forms ($r > 0.99$).

The dosage form is a dispersion of tacrolimus, dissolved in propylene carbonate, in a continuous phase consisting of mineral oil, parrafin, white wax, and white petrolatum. The critical parameter, droplet size distribution, is dependent on the type of manufacturing equipment used to disperse the solution of drug substance in the ointment base.

In vitro release testing was used to compare the release rates of the two products, to ensure that no differences in product performance would arise from the differences in microscopic structures.

Comparison of the release rates for 0.3% ointments made in pilot-scale vs. production-scale equipment showed that the two rates were not significantly different (ANCOVA, $p > 0.05$), and were linear and directly proportional to the square root of time ($r > 0.99$). These results indicate that there are no differences in the dosage forms manufactured using different equipment.