had higher spleen weights (23% and 59% at 10 and 100 mg/kg, respectively), and females receiving this dose had higher liver weights (31%).

Histopathology: Foamy macrophages (finely vacuolated cells) were observed in the following organs from animals treated with the 100 mg/kg dose for either 13 or 26 weeks: choroid plexus of the brain, adrenals, mesenteric and mandibular lymph nodes, lamina propria of the urinary bladder and endometrial stromal mucosa of the uterus and spleen. In addition, minimal vacuolation in the epithelial cells of the choroid plexus was also observed. The changes were not completely resolved at the end of the 13-week recovery period. At the end of the 13 or 26 weeks treatment period, foamy macrophages were observed at the injection sites of animals treated with 10 and 100 mg/kg doses. This was reversible in the recovery group animals treated for 13 weeks, and those receiving the 10 mg/kg dose for 26 weeks, but not in those receiving the 100 mg/kg dose for 26 weeks. The incidences of foamy macrophages in different groups of male and female animals are summarized in the Table below.

Organ/Findings	**Control**		**10 mg/kg/week**		**100 mg/kg/week**	
(26 Weeks treatment)	**Male**	**Female**	**Male**	**Female**	**Male**	**Female**
Adrenal						
– Foamy macrophages	0/3	0/3	0/4	0/4	3/4	3/4
Brain						
- Foamy macrophages	0/3	0/3	0/4	0/4	2/4	4/4
Mesenteric lymph nodes						
- Foamy macrophages	0/3	0/3	0/4	0/4	0/4	4/4
Spleen						
- Foamy macrophages	0/3	0/3	0/4	0/4	0/4	2/4
Urinary bladder						
- Foamy macrophages	0/3	0/3	0/4	0/4	1/4	3/4
Uterus						
- Foamy macrophages	0/3	0/3	0/4	0/4	0/4	4/4
Injection sites						
- Foamy macrophages (upper left)	0/3	0/3	2/4	4/4	3/4	4/4
- Foamy macrophages (lower left)	0/3	0/3	0/4	1/4	3/4	3/4
- Foamy macrophages (upper right)	0/3	0/3	2/4	4/4	4/4	3/4
- Foamy macrophages (lower right)	0/3	0/3	2/4	2/4	3/4	3/4
13 Weeks treatment						
Adrenal						
– Foamy macrophages	0/3	0/3	0/4	0/5	2/4	0/4
Brain						
- Foamy macrophages	0/3	0/3	0/4	0/5	2/4	0/4
Mesenteric lymph nodes						
- Foamy macrophages	0/3	0/3	0/4	0/5	2/4	1/4

Urinary bladder - Foamy macrophages	0/3	0/3	0/4	0/5	1/4	0/4
Uterus - Foamy macrophages	0/3	0/3	0/4	0/5	0/4	2/4
Injection sites - Foamy macrophages (upper left)	0/3	0/3	1/4	3/5	3/4	2/4
- Foamy macrophages (lower left)	0/3	0/3	1/4	0/5	2/5	4/4
- Foamy macrophages (upper right)	0/3	0/3	1/4	3/5	4/4	4/4
- Foamy macrophages (lower right)	0/3	0/3	1/4	4/5	3/4	4/4

Toxicokinetics: Two female monkeys receiving the 10 mg/kg dose had detectable antibodies in week 9, and these animals were terminated in week 13. Plasma concentrations of CDP870 increased with increasing doses, and the increase was dose-proportional. Following the first injection, mean plasma concentrations at 48 hours were 93.74 and 903.31 μg/ml at 10 and 100 mg/kg doses, respectively. These concentrations declined to 63.96 and 587.13 μg/ml, respectively by day 7. The steady state concentrations were reached by the 5th injection, and the plasma concentrations following the 5th injection were 190.98 and 1856.97 μg/ml at 10 and 100 mg/kg doses, respectively. The half-life of elimination following 13 or 26 weeks treatment ranged from 8.5 to 10.3 days. Plasma pharmacokinetic parameters for CDP870 of individual animal are shown in the Table below.

TABLE 3
PAGE 1 OF 3
INDIVIDUAL PHARMACOKINETIC PARAMETER ESTIMATES - CDP870 PLASMA CONCENTRATIONS (μg/mL)

DOSE GROUP	LENGTH OF TIME IN THE STUDY	GENDER	MONKEY NUMBER	C max (μg/mL)	T max (weeks)	ß (h^-1)	T1/2 (hours)	T1/2 (days)	AUC (0-t) (μg.h/mL)	AUC (0-inf.) (μg.h/mL)
CDP870 10 MG/KG	13 WEEKS	FEMALE	2234	255.8	4.3	.	.	.	244577.2	.
			2235	281.3	8.3	.	.	.	261173.8	.
			2236	203.0	11.3	.	.	.	265740.2	.
		MALE	2221	246.6	11.3	.	.	.	333113.1	.
			2222	259.3	11.3	.	.	.	288310.4	.
			2223	287.7	11.3	.	.	.	331797.5	.
			2224	214.6	4.3	.	.	.	298340.1	.
	18 WEEKS	MALE	2231	308.5	11.3	.	.	.	424570.6	.
	13+13 WEEKS	FEMALE	2238	218.5	4.3	.	.	.	279539.2	.
			2240	181.4	4.3	.	.	.	226540.3	.
		MALE	2225	236.9	4.3	.	.	.	332550.7	.
			2226	190.9	8.3	.	.	.	323349.1	.
	26 WEEKS	FEMALE	2241	186.5	25.3	.	.	.	507399.3	.
			2242	197.6	19.3	.	.	.	489898.5	.
			2243	207.3	19.3	.	.	.	544955.6	.
			2244	199.4	23.3	.	.	.	447345.0	.
		MALE	2227	249.4	19.3	.	.	.	507780.6	.
			2228	275.7	23.3	.	.	.	664837.8	.
			2229	274.3	19.3	.	.	.	626825.4	.
			2230	229.7	23.3	.	.	.	512879.4	.

Note: The limit of quantification for measuring CDP870 plasma concentration was set to —
Half-life has been calculated for monkeys with three data points in the elimination phase.
All observations for monkeys 2237 and 2239 of the 10 mg/kg CDP870 group have been excluded from the summary statistics.

TABLE 3
PAGE 2 OF 3
INDIVIDUAL PHARMACOKINETIC PARAMETER ESTIMATES - CDP870 PLASMA CONCENTRATIONS (µg/mL)

DOSE GROUP	LENGTH OF TIME IN THE STUDY	GENDER	MONKEY NUMBER	C max (µg/mL)	T max (weeks)	ß (h^-1)	T1/2 (hours)	T1/2 (days)	AUC (0-t) (µg.h/mL)	AUC (0-inf.) (µg.h/mL)
CDP870 10 MG/KG	26+13 WEEKS	FEMALE	2245	284.7	23.3	.	.	.	554071.4	.
			2246	240.4	4.3	.	.	.	612784.6	.
		MALE	2232	219.1	19.3	.	.	.	644097.2	.
			2233	246.2	23.3	.	.	.	624714.5	.
CDP870 100 MG/KG	13 WEEKS	FEMALE	2260	3140.7	4.3	.	.	.	3572407.3	.
			2261	2575.1	4.3	.	.	.	3036407.3	.
			2262	2523.4	4.3	.	.	.	2984795.5	.
			2263	1983.9	4.3	.	.	.	2661998.0	.
		MALE	2247	3334.4	4.3	.	.	.	3734474.3	.
			2248	2389.5	4.3	.	.	.	2606644.0	.
			2249	2756.3	11.3	.	.	.	3486611.7	.
			2250	2095.8	11.3	.	.	.	3061769.1	.
	18 WEEKS	FEMALE	2266	1775.1	11.3	.	.	.	3230298.8	.
		MALE	2257	1433.4	11.3	.	.	.	2666982.7	.
	13+13 WEEKS	FEMALE	2264	1646.6	8.3	.	.	.	2292340.7	.
			2265	1668.7	4.3	0.0034	205.5	8.6	2438834.8	2439220.1
		MALE	2251	1490.6	4.3	0.0028	246.5	10.3	2619311.5	2620449.5
			2252	2431.5	11.3	0.0029	243.0	10.1	3260077.4	3261094.0

Note: The limit of quantification for measuring CDP870 plasma concentration was set tc [redacted] µg/mL.
Half-life has been calculated for monkeys with three data points in the elimination phase.
All observations for monkeys 2237 and 2239 of the 10 mg/kg CDP870 group have been excluded from the summary statistics.
Plasma concentration at 48 hours and 7 days post-1st injection were excluded from the summary statistics therefore AUC was not calculated for monkey 2254 of the 100 mg/kg.

TABLE 3
PAGE 3 OF 3
INDIVIDUAL PHARMACOKINETIC PARAMETER ESTIMATES - CDP870 PLASMA CONCENTRATIONS (µg/mL)

DOSE GROUP	LENGTH OF TIME IN THE STUDY	GENDER	MONKEY NUMBER	C max (µg/mL)	T max (weeks)	ß (h^-1)	T1/2 (hours)	T1/2 (days)	AUC (0-t) (µg.h/mL)	AUC (0-inf.) (µg.h/mL)
CDP870 100 MG/KG	26 WEEKS	FEMALE	2267	2228.6	25.3	.	.	.	5159575.8	.
			2268	2254.6	23.3	.	.	.	4570142.3	.
			2269	2633.9	4.3	.	.	.	5299598.8	.
			2270	2047.9	23.3	.	.	.	5547985.0	.
		MALE	2253	2256.1	25.4	.	.	.	5479281.3	.
			2255	1897.1	19.3	.	.	.	5203207.0	.
			2256	2149.4	25.4	.	.	.	4781649.7	.
	26+13 WEEKS	FEMALE	2271	2058.4	19.3	0.0030	234.7	9.8	4677604.8	4677977.3
			2272	2494.1	4.3	0.0033	208.3	8.7	6049574.7	6049935.4
		MALE	2258	2183.4	23.3	0.0034	205.0	8.5	5022400.7	5022726.0
			2259	1906.1	23.3	0.0032	214.6	8.9	4854422.2	4854886.6

Note: The limit of quantification for measuring CDP870 plasma concentration was set to [redacted].
Half-life has been calculated for monkeys with three data points in the elimination phase.
Plasma concentration at 48 hours and 7 days post-1st injection were excluded from the summary statistics therefore AUC was not calculated for monkey 2254 of the 100 mg/kg.

Anti-CDP870 antibody levels of the control and treatment groups at different times are shown in the Table below.

TABLE 2
PAGE 1 OF 2

ANTI-CDP870 LEVELS (units/mL) - SUMMARY STATISTICS

SCHEDULED VISIT		CDP870 DOSE (mg/kg)		
		CONTROL	10	100
PRE-1ST INJECTION	MEAN	0.300	0.300	0.300
	95% CI	0.3000, 0.3000	0.3000, 0.3000	0.3000, 0.3000
	N	6	7	8
7 DAYS POST-1ST INJECTION	MEAN	0.300	0.300	0.300
	95% CI	0.3000, 0.3000	0.3000, 0.3000	0.3000, 0.3000
	N	6	7	8
PRE-5TH INJECTION	MEAN	0.300	0.300	0.300
	95% CI	0.3000, 0.3000	0.3000, 0.3000	0.3000, 0.3000
	N	6	7	8
7 DAYS POST-5TH INJECTION	MEAN	0.300	0.300	0.300
	95% CI	0.3000, 0.3000	0.3000, 0.3000	0.3000, 0.3000
	N	6	7	8
PRE-9TH INJECTION	MEAN	0.300	0.300	0.300
	95% CI	0.3000, 0.3000	0.3000, 0.3000	0.3000, 0.3000
	N	6	7	8
7 DAYS POST-9TH INJECTION	MEAN	0.300	0.300	0.300
	95% CI	0.3000, 0.3000	0.3000, 0.3000	0.3000, 0.3000
	N	6	7	8
PRE-12TH INJECTION	MEAN	0.300	0.300	0.300
	95% CI	0.3000, 0.3000	0.3000, 0.3000	0.3000, 0.3000
	N	6	7	8

Note: The geometric mean and 95% confidence interval have been presented.
The limit of quantification was set to [redacted] units/mL.
All observations for monkeys 2237 and 2239 of the 10 mg/kg CDP870 group have been excluded from the summary statistics.

ANTI-CDP870 LEVELS (units/mL) - SUMMARY STATISTICS

SCHEDULED VISIT		CDP870 DOSE (mg/kg)		
		CONTROL	10	100
7 DAYS POST-12TH INJECTION	MEAN	0.300	0.324	0.304
	95% CI	0.3000, 0.3000	0.2777, 0.3786	0.2952, 0.3123
	N	6	7	8
NECROPSIS	MEAN	0.300	0.300	0.300
	95% CI	0.3000, 0.3000	0.3000, 0.3000	0.3000, 0.3000
	N	6	7	8

Note: The geometric mean and 95% confidence interval have been presented.
The limit of quantification was set to [redacted] units/mL.
All observations for monkeys 2237 and 2239 of the 10 mg/kg CDP870 group have been excluded from the summary statistics.

Summary: In the 13/26 –week subcutaneous toxicity study with CDP870 in cynomolgus monkeys, the drug was administered to groups of animals at doses of 0, 10 and 100 mg/kg, once weekly. At the end of the 13 or 26 weeks treatment period, 2 animals/sex/group were left untreated for a 13-week recovery period. There were no treatment-related changes in clinical signs, body weight, ECG, gross pathology, histopathology or clinical chemistry parameters in any group. Foamy macrophages in different organs were observed in animals receiving the 100 mg/kg weekly dose for 13 or 26 weeks. The changes were not completely reversible at the end of the 13-week recovery period. Foamy macrophages at the injection sites were observed in animals receiving both 10 and 100 mg/kg doses. The presence of foamy macrophages at the injection sites was reversible in the 10 mg/kg dose group but not in the 100 mg/kg dose group. Anti-CDP870 antibodies were detected in week 9 in two females receiving the 10 mg/kg dose, and these animals were sacrificed. Six more animals had low levels of anti-CDP870 antibodies at the end of the dosing period. The target organs of toxicity were the injection site, brain, adrenals,

mesenteric and mandibular lymph nodes, urinary bladder, uterus and spleen. The 10 mg/kg weekly dose was the no effect dose.

Study Title: 52-Week Study with a Recovery Period in Cynomolgus Monkeys To Examine the Effects of CDP870 on Hematological and Morphological Parameters following Repeat Subcutaneous Dosing.

Study Report No: 506771

Conducting laboratory and location: [redacted]
Date of study initiation: February 18, 2004

Date of study completion: April 07, 2006
GLP compliance: Yes
QA- Report Yes ( ) No ( ): Yes
Methods: The potential effects of CDP870 on the lymphoid tissue/lymphocyte subsets were examined in cynomolgus monkeys after once weekly subcutaneous administration for 52 weeks.

Dosing:

- **species/strain:** Cynomolgus monkey (*Macaca fascicularis*)
- **#/sex/group or time point:** 8 to 10 animals/sex/group
- **age:** 17 to 27 months old.
- **weight:** 1.9 to 3.8 kg.
- **Satellite groups used for toxicokinetics or recovery:** Half of the animals were sacrificed at the end of the 52-week dosing period, and the remaining animals were placed on a 26-week reversibility.
- **Dosage groups in administered units:** 50 and 100 mg/kg/week. The sponsor stated that the doses were based on results from earlier studies in monkeys, and the dose selection was in agreement with the FDA.
- **Route, form, volume, and infusion rate:** CDP870 was administered by subcutaneous injections once weekly at a dosing volume of 0.5 ml/kg.

Drug, lot#, radiolabel, and % purity: CDP870, Batch no. PCE003; purity, 100%.
Formulation/vehicle: Solutions of the lyophilized drug were prepared by adding water for injections, and dilutions were made using the vehicle containing [redacted] sucrose (w/v) and [redacted] Tween (w/v).
Observations and times:

- **Clinical signs:** Animals were observed twice daily for clinical signs of toxicity and mortality.
- **Body weights:** Body weight of the animals was measured once weekly during the dosing period.
- **Food and water consumption:** Food and water consumptions were not measured.
- **Ophthalmoscopy:** Ophthalmoscopic examinations of all animals were conducted before initiation of dosing, and during weeks 26 and 52.

- **Hematology:** Blood samples for measurement of hematology parameters were collected from all animals before initiation of dosing and during weeks 12, 24, 41 and 52 of the treatment period.

- **Clinical chemistry:** Blood samples for clinical chemistry investigations were collected before initiation of dosing and during weeks 12, 24, 41 and 52 of the treatment period.

- **Immunological investigations:** Whole blood samples (1 ml) for flow cytometry analysis were obtained weekly from weeks -12 to -1, and on weeks 1, 2, 3, 4, 8, 12, 16, 20, 24, 28, 32, 36, 40, 44, 48 and 52 of the treatment period. The following CD markers were used in the study: CD3, CD4, CD8, CD16, CD20, CD21, CD23 and CD40.

- **Gross pathology:** At the end of the treatment period, half of the animals were sacrificed and complete necropsies performed. The remaining half of the animals underwent a 26-week treatment-free recovery period. Animals allocated in the main and recovery groups are shown in the Table below.

Group	Treatment (mg/kg/week)	Animal			
		Main Phase		Recovery Phase	
		Males	Females	Males	Females
1	0	5-8	27-30	1-4	31-34
2	50	9-12	35-38	13-16	39,40,42
3	100	22-26	43-46,48	17-21	49-52

Premature decedents, Animal 47F (Week 3) Animal 41F (Week 44).

- **Organs weighed:** The weights of the following organs were determined at necropsy: adrenals, brain, heart, kidneys, liver, lungs, ovaries, pancreas, pituitary gland, prostate, spleen, submaxillary glands, testes, thymus, thyroid, uterus, and cervix plus oviduct.

- **Histopathology:** The following organs and tissues from all animals were examined histologically: adrenal, brain, ileum, injection sites, mesenteric lymph nodes, spleen, sternum, submandibular lymph node, thymus, urinary bladder, uterus and cervix plus oviduct.

- **Toxicokinetics:** Blood samples for measurement of the plasma concentrations of CDP870 was collected pre-dose, and approximately 48 hours and 7 days (before the next dose) after the 1st, 6th, 12th, 24th, 38th and 50th dose. An additional sample was also taken 7 days after the last dose. Blood samples from the recovery animals were collected 7 days after the last dose, and in recovery weeks 4, 8, 12, 16, 20, 24 and 26. Plasma concentrations of CDP870 were measured using an ELISA method. Blood samples were also analyzed for antibodies to CDP870.

Results:

- **Clinical signs:** Soft/liquid feces were observed in females receiving 50 and 100 mg/kg/week doses during most part of the dosing period. Treatment group females also had higher incidences of hair loss at the back, head and shoulders.

- **Mortality:** A female monkey (#47F) receiving the 100 mg/kg/week dose was found dead on day 18 of the study. At necropsy, the abdomen was swollen due to distended intestines, and the tongue was protruding from the mouth. A 50 mg/kg/week dose group female was sacrificed for humane reasons on day 304 (week 44) of the study. In the weeks before sacrifice, this animal showed body weight loss, was subdued and had swollen abdomen. Pathological findings included

mild to moderate inflammation of the colon and rectum, and an absent thymus indicating severe thymic atrophy.

- **Body weights:** The mean body weights of the control male and female animals were 3.1±0.5 and 2.5±0.2 kg on the day before initiation of dosing, and 4.4±1.0 and 3.6±0.7 kg at the end of the dosing period, respectively. The body weights of the animals were not affected by treatment with CDP870.

- **Ophthalmoscopy:** No treatment-related ophthalmologic changes were observed in any group.

- **Hematology:** Activated partial thromboplastin times (APTT) were increased (up to 33%) in weeks 12, 24, 41 and 52 of treatment in both males and females receiving CDP870 when compared to the control group (The APTT values of control males and females before initiation of dosing were 30±3 and 32±7 seconds, respectively). However, the increases were not dose-dependent. Males receiving 50 and 100 mg/kg/week doses had slight decreases in RBC (control, $6.76x10^{12}$; up to 7% increase), hemoglobin (control, 12.9±0.6 g/dl; up to 8% increase) and hematocrit (control, 0.465±0.02 L/L; up to 7% increase) levels throughout the treatment period.

- **Clinical chemistry:** Females receiving CDP870 had decreased lactase dehydrogenase (LDH) levels in weeks 12 (control, 635±78 IU/L; 16.7% and 27.7% increases at 50 and 100 mg/kg, respectively), 24 (control, 361±83 IU/L; 22.4% and 23% increases at 50 and 100 mg/kg, respectively), 41 (control, 295±32 IU/L; 17% and 21.4% increases at 50 and 100 mg/kg, respectively) and 52 (control, 283±34 IU/L; 25.4% and 19.4% increases at 50 and 100 mg/kg, respectively), when compared with the control. Treatment group females had lower cholesterol levels during the dosing period. The cholesterol level of control females before initiation of dosing was 3.7±0.4 mmol/L, and there were 17% to 33% decreases during the dosing period.

- **Immunological investigation:** During the treatment period, the different CD marker sets absolute counts were relatively stable until between weeks 28-32, when there was a non-specific increase in the different lymphocyte counts in all groups including controls. Similarly, the number of mature B cells showed a gradual increase over time in all groups including the control.

- **Organ weight:** The absolute (23.6%) and relative (19.8%) weights of the liver were slightly higher in males receiving the 100 mg/kg/week dose and females receiving 50 (12.7%, absolute) and 100 (13.8%, absolute) mg/kg/week doses.

- **Gross pathology:** No treatment-related gross pathological changes were observed in any group. Occasional reddening or dark foci were observed at the injection sites of animals from all groups.

- **Histopathology:** Animals treated with CDP870 at 100 mg/kg/week showed vacuolation in several organs, including sinusoidal histiocytes, submandibular and mesenteric lymph nodes, splenic red pulp, zona reticularis of the adrenal gland, lamina propria of fallopian tube, uterine body and cervix, choroid plexus of the brain and the subcutaneous tissue of the injection site. In the lamina propria of some organs such as the uterus and brain, as well as the injection site, the vacuolation was evident as large foamy macrophages. In animals treated with the 50 mg/kg/week dose, a reduced incidence of vacuolation was present in mesenteric lymph node, splenic red pulp, fallopian tube and uterine body, choroid plexus of the brain and the injection site. There was a minimal or mild chronic subcutaneous inflammation at the injection sites of most animals treated with CDP870.

Histopathological findings for male and female monkeys are shown in the Table below.

Organ/Lesion	Male			Female		
	Control	50 mg/kg	100 mg/kg	Control	50 mg/kg	100 mg/kg
Lymph node (Mandibular)						
-Vacuolation, histiocytic	0/4	0/4	2/5	0/4	0/4	1/5
Lymph node (Mesenteric)						
-Vacuolation, histiocytic	0/4	1/4	4/5	0/4	1/4	4/5
Spleen						
-vacuolation, red pulp	0/4	0/4	4/5	0/4	1/4	3/5
Adrenal gland						
-vacuolation, zona reticularis	0/4	0/4	4/5	1/4	0/4	3/5
Uterus						
-vacuolation, fallopian tube	--	--	--	0/4	1/4	3/5
-vacuolation, lamina propria	--	--	--	0/4	2/4	4/5
-vacuolation, cervix, lamina propria	--	--	--	0/4	0/4	1/5
Brain						
-Vacuolation, choroid plexus, epithelial	0/4	2/4	2/5	0/4	3/4	0/5
-Lymphoid follicular hyperplasia,choroid plexus	0/4	0/4	1/5	0/4	0/4	0/5
Injection sites						
-vacuolation, histiocytic	0/4	1/4	3/5	0/4	3/4	2/5
-inflammation, subcutaneous, chronic	0/4	3/4	4/5	1/4	4/4	3/5

Following a 26-week recovery period, vacuolation was still present in mesenteric lymph nodes, the zona reticularis of the adrenal gland, the lamina propria of fallopian tube, uterine body and cervix, and the lamina propria and epithelium of the choroid plexus of the brain in animals administered CDP870 at 50 and 100 mg/kg/week doses. There were foamy macrophages in the spleen of one animal. Histiocytic vacuolation at the injection sites and mandibular lymph nodes were not present in the recovery animals. Histopathological findings in the recovery animals are shown in the Table below.

Organ/Lesion	Male			Female		
	Control	50 mg/kg	100 mg/kg	Control	50 mg/kg	100 mg/kg
Lymph node (Mesenteric)						
-Vacuolation, histiocytic	0/4	1/4	3/5	0/4	0/3	1/4
Spleen						
-foamy macrophages, red pulp, minimal	0/4	0/4	1/5	0/4	0/3	0/4
Adrenal gland						
-vacuolation, zona reticularis	0/4	20/4	4/5	0/4	0/3	3/4
Uterus						
-vacuolation, fallopian tube	--	--	--	0/4	2/3	4/4
-vacuolation, lamina propria	--	--	--	0/4	2/3	3/4
-vacuolation, cervix, lamina propria	--	--	--	0/4	1/3	3/4
Brain						
-Vacuolation, choroid plexus, epithelial	0/4	2/4	4/5	0/4	1/3	0/4

Toxicokinetics:

The plasma CDP870 concentrations increased with increasing dose in both male and female monkeys. No differences in the C_{max} values were observed between males and females. The mean C_{max} values at 50 and 100 mg/kg doses 924 and 1692 µg/ml, respectively. The median T_{max} values at 50 and 100 mg/kg doses were 212 and 163 days, respectively. The systemic exposures at 50 and 100 mg/kg doses, as measured by AUC at steady state over a 7-day dosing interval between days 343 and 350, were 4423 and 7202 µg.day/ml, respectively. Attainment of steady state occurred at approximately 42 days. The pharmacokinetic parameters of individual animals treated with 50 and 100 mg/kg/week doses are shown in the Tables below.

Table 3 Individual Pharmacokinetic Parameters 50 mg/kg administration

Treatment	Subject	Cmax (µg/ml)	AUCt (µg.day/ml)	AUCt (µg.hr/ml)
50mg/kg	009-2M	942.01	180386.65	4329279.65
	010-2M	1064.64	158537.77	3804906.42
	011-2M	873.86	179233.56	4301605.5
	012-2M	778.00	152124.18	3650980.28
	013-2M	1061.35	215683.70	5176408.9
	014-2M	887.46	161010.49	3864251.74
	015-2M	694.87	149601.43	3590434.26
	016-2M	960.93	242246.71	5813921.05
	035-2F	886.75	196992.85	4727828.33
	036-2F	921.72	187225.34	4493408.16
	037-2F	962.52	183890.47	4413371.36
	038-2F	909.76	148570.55	3565693.14
	039-2F	1034.43	261008.00	6264192.1
	040-2F	954.99	212422.74	5098145.65
	041-2F	953.44	100321.53+	2407716.83+
	042-2F	988.47	195435.86	4690460.75
	N	16	16	16
	Geometric Mean	924.694	178632.822	4287187.724
	SD of the Logs	0.1094	0.2269	0.2269
	CV% Geometric Mean	11.0	23.0	23.0

Table 4 Individual Pharmacokinetic Parameters 100 mg/kg administration

Treatment	Subject	Cmax (µg/ml)	AUCt (µg.day/ml)	AUCt (µg.hr/ml)
100mg/kg	017-3M	2063.16	461271.92	11070526.19
	018-3M	1466.13	339780.36	8154728.54
	019-3M	1419.12	251533.29	6036798.9
	020-3M	1540.83	326449.15	7834779.53
	021-3M	2176.11	391851.87	9404444.94
	022-3M	1774.70	283079.75	6793914.06
	023-3M	1670.41	326623.95	7838974.88
	024-3M	1713.79	333582.91	8005989.73
	025-3M	1574.85	294671.14	7072107.4
	026-3M	2022.70	374282.10	8982770.37
	043-3F	2101.05	279546.82	6709123.74
	044-3F	1499.62	217636.04	5223265.07
	045-3F	1747.76	264844.02	6356256.37
	046-3F	1128.44	211531.62	5076758.97
	048-3F	1453.94	230688.58	5536525.86
	049-3F	1913.47	332575.71	7981817.09
	050-3F	2219.00	312262.31	7494295.4
	051-3F	1435.38	257982.01	6191568.35
	052-3F	1700.60	218313.03	5239512.74
	N	19	19	19
	Geometric Mean	1691.817	293944.507	7054668.173
	SD of the Logs	0.1776	0.2140	0.214
	CV% Geometric Mean	17.9	21.6	21.6

Low levels antibodies to CDP870 were detected in two animals receiving the 50 mg/kg/week dose (0.72 and 0.62 units/ml, respectively). The PK profile of these animals was similar to other animals in this group. One male monkey (#10M) receiving the 50 mg/kg/week dose showed a marked increase in clearance of CDP870 over time indicating the possible influence of anti-CDP870, although antibodies were not detected in this animal.

Key Study Findings: In a 52-week subcutaneous toxicology study with CDP870 in cynomolgus monkeys, the drug was administered at 50 and 100 mg/kg doses, once weekly. Although, the sponsor stated that half of the animals were left untreated for a 26-week recovery period, no data were provided for the recovery groups in this interim study report. An increase in the activated partial thromboplastin time (APTT) was observed in males and females receiving CDP870. CDP870 had no significant effect on the immune system of the animals. Animals treated with CDP870 at 100 mg/kg/week showed vacuolation in several organs, including sinusoidal histiocytes, submandibular and mesenteric lymph nodes, splenic red pulp, zona reticularis of the adrenal gland, lamina propria of fallopian tube, uterine body and cervix, choroid plexus of the brain and the subcutaneous tissue of the injection site. Vacuolation was also observed in some organs at the 50 mg/kg/week dose. At the end of the 26-week recovery period, vacuolation was still present in most organs, although with a less severity.
Following s.c. administration to monkeys, the plasma exposure levels of CDP870 increased with increasing dose with no differences between males and females. Antibodies to CDP870 were detected in two animals receiving the 50 mg/kg dose.

2.6.6.1 Overall toxicology summary

Toxicology studies with CDP870 were conducted in monkeys following administration of single and multiple doses. Intravenous administration of single doses up to 870 mg/kg was well-tolerated without any treatment-related adverse effects. In a 28-day i.v. toxicity study in monkeys, decreased hemoglobin, RBC and packed cell volume was observed immediately after administration of 50, 100 and 400 mg/kg weekly doses. Increased WBC levels were observed in male and female monkeys receiving 10 and 100 mg/kg i.v. doses for 13 and 26 weeks. Histiocytic vacuolation in the hemolymphoreticular tissues (splenic red pulp, medullary sinuses of the mandibular and mesenteric lymph nodes, bone marrow, thymus) were observed in animals treated with the 400 mg/kg dose for 28 days. Vacuolation (foamy macrophages) was also observed in different organs (choroid plexus, adrenals, mesenteric and mandibular lymph nodes, lamina propria of the urinary bladder and endometrial stromal mucosa of the uterus and spleen) in monkeys receiving a 100 mg/kg weekly dose for 13, 26 or 52 weeks. Vacuolation of the hemolymphoreticular tissues may be related to the pharmacological effects of the drug. An increase in the activated partial thromboplastin time (APTT) was observed in monkeys receiving 50 and 100 mg/kg doses. A similar increase in the APTT was also observed in an *ex vivo* study with monkey blood. However, no effects on APTT were observed in animals receiving the drug for 13 or 26 weeks.

2.6.6.4 **Genetic toxicology**

Descriptive Title: CDP870: Reverse Mutation in Four Histidine-Requiring Strains of Salmonella Typhimurium and One Tryptophan-Requiring Strain of Escherichia Coli.

Sponsor's ID # Study No. ARLE05K2706 (Study Report # 40000945)

Conducting Laboratory: [redacted]

Date of Study Initiation/completion: March 30, 1999/January 08, 1999

GLP Compliance: Yes

Drug Lot Number: CDP870; Lot No. 97LM202P

Study Endpoint: Mutagenesis

METHODOLOGY:

Strains/Species/Cell line: *Salmonella typhimurium* strains TA100, TA98, TA1535 and TA1537, and *Escherichia coli* strain WP2uvrA

Dose Selection Criteria: An initial dose-ranging experiment was carried out in strain TA100 only, using final concentrations of CDP870 ranging from 7.995 to 4997 µg/plate (7.995, 39.97, 199.9, 999.4 and 4997 µg/plate). The maximum dose was selected as the closest approximation to 5000 µg/plate that could be achieved without volume additions. Triplicate samples were incubated with CDP870 without metabolic activation at 37^0c for 3 days. Following incubation, the plates were examined for evidence of toxicity, and where possible, revertant colonies were counted.

Test Agent Stability Considerations: It was stated that the solutions of the test agent were used within 5 hours of removal from the storage condition, and it was stable during this period.

Metabolic Activation System: Rat liver microsomal S9 fraction prepared from male Sprague-Dawley rats treated with Aroclor 1254 was obtained from [redacted].

Controls: Acetate buffer (pH 5.5) was used as a negative control. The following agents were used as positive controls in the absence of metabolic activation: 2-nitrofluorene (2NF, for TA98), sodium azide (NaN3, for TA100 and TA1535), 9-aminoacridine (AAC, for TA1537) and 4-nitroquinolone 1-oxide (NQO, for WP2 uvrA). 2-aminoanthracene (AAN) was used as a positive control in the presence of metabolic activation.

Exposure conditions: The tests were performed using the plate incubation method with or without S9 mix. The bacterial suspensions (with the test substance or controls in absence or presence of metabolic activation) were shake-cultured for 60 minutes at 37^0C and top agar was added to the mixture. Plating of these treatments then proceeded as for the normal plate incorporation procedure.

Analysis: The numbers of revertant colonies were counted automatically using a — Colony Counter or manually. The mean of the number of revertant colonies at each concentration was determined and compared with the negative controls.

Criteria for Positive Results: The study was considered positive if there was a significant ($p \leq 0.01$; Dunnett's test) and dose related reproducible increase in the number of revertant colonies in the treatment groups when compared with the negative control.

RESULTS

Study Validity: the study was considered valid if the mean number of revertant colonies in the negative controls fell within the normal ranges from the conducting laboratory, the positive controls induced a clear increase in the number of revertant colonies, and no more than 5% of the plates were lost due to contamination or other reasons. Based on these criteria, the study was valid.

Study Outcome: CDP870 was soluble in the culture medium at concentrations up to 4997 µg/plate, and no cytotoxicities were observed for any strain. CDP870 did not cause an increase in the number of revertant colonies in any of the strains of *Salmonella typhimurium* or *E. coli* in the absence or presence of metabolic activation under the conditions of the experiment. Thus, CDP870 was not mutagenic in this assay. The number of mean revertant colonies in the main study for different strains in the absence or presence of metabolic activation is shown in the Table below.

CDP 870: summary of mean revertant colonies (-S-9) - Experiment 1

Substance	Dose Level µg/plate	TA98	TA100	TA1535	TA1537	WP2 uvrA
		Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
PLACEBO BUFFER SOLN.	240 µl	30 ± 7	115 ± 8	11 ± 7	6 ± 2	23 ± 6
Untreated	0 µl	-	105 ± 4	-	-	-
CDP 870	7.995	32 ± 6	120 ± 15	10 ± 3	6 ± 1	25 ± 8
	39.97	35 ± 7	114 ± 17	10 ± 4	6 ± 3	21 ± 4
	199.9	30 ± 6	134 ± 16	11 ± 3	6 ± 3	21 ± 7
	999.4	32 ± 3	108 ± 6	9 ± 4	5 ± 3	24 ± 4
	4997	24 ± 2	105 ± 6	11 ± 6	5 ± 1	19 ± 6
Positive controls	Compound	2NF	NaN3	NaN3	AAC	NQO
	Dose Level	5 µg	2 µg	2 µg	50 µg	2 µg
	Mean ± SD	651 ± 8	499 ± 22	216 ± 19	264 ± 3	433 ± 36

SD Standard deviation

2NF 2-Nitrofluorene
NaN3 Sodium azide
AAC 9-Aminoacridine
NQO 4-Nitroquinoline 1-oxide

CDP 870: summary of mean revertant colonies (+S-9) - Experiment 1

Substance	Dose Level µg/plate	TA98	TA100	TA1535	TA1537	WP2 uvrA
		Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
PLACEBO BUFFER SOLN.	240 µl	42 ± 13	147 ± 10	14 ± 2	9 ± 3	27 ± 5
Untreated	0 µl	-	144 ± 10	-	-	-
CDP 870	7.995	47 ± 12	131 ± 12	14 ± 4	9 ± 2	24 ± 3
	39.97	44 ± 1	143 ± 5	17 ± 4	7 ± 1	25 ± 6
	199.9	40 ± 9	155 ± 16	15 ± 4	7 ± 4	26 ± 5
	999.4	46 ± 12	152 ± 6	13 ± 1	8 ± 3	28 ± 6
	4997	45 ± 5	147 ± 22	14 ± 6	6 ± 3	25 ± 8
Positive controls	Compound	AAN	AAN	-	-	AAN
	Dose Level	5 µg	5 µg	-	-	10 µg
	Mean ± SD	583 ± 53	846 ± 62	-	-	70 ± 23

SD Standard deviation

AAN 2-Aminoanthracene

SUMMARY:

CDP870 did not cause a significant increase in the number of revertant colonies for any of the tester strains in the absence or presence of metabolic activation. Thus, the compound was not mutagenic in the test systems under the experimental conditions.

Study Title: **CDP870: Induction of Chromosome Aberrations in Cultured Human Peripheral Blood Lymphocytes.**

Study Report No. 40000980

Conducting Laboratory: [redacted]

Date of Study Initiation/completion: March 30, 1998/ March 24, 1999

GLP Compliance: Yes

Drug Lot Number: CDP870, Lot No. 97LM202P; purity, >95%.

Study Endpoint: Chromosomal aberration.

METHODOLOGY:

Strains/Species/Cell line: Human peripheral blood lymphocytes obtained from healthy donors were used in the study.

Dose Selection Criteria: The highest dose for chromosome aberration was selected on the basis of mitotic inhibition (the highest dose should produce at least 50% mitotic inhibition) and precipitation of the test agent. Slides from cultures from heavily precipitating doses were checked to confirm that the presence of precipitate did not preclude analysis.

Test Agent Stability Considerations: It was stated that CDP870 was used within 2 hours of dilution from the storage sample, and it was stable during this period.

Metabolic Activation System: Rat liver microsomal S9 fraction from Aroclor 1254-treated male Sprague-Dawley rats ([redacted]) was used as a metabolic activator.

Controls: Acetate buffer (pH 5.5) was used as a negative control. 4-nitroquinoline 1-oxide (NQO) was used a positive control in the absence of metabolic activation, and cyclophosphamide (CPA) was used as a positive control in the presence of metabolic activation.

Exposure conditions: Quadruplicate cultures for each of the treatment regimens were treated with the solvent, positive control or the test agent in the presence or absence of S9 mix. The mixtures were incubated for 20+0 hours in the absence of metabolic activation and 3+17 hours in the absence and presence of metabolic activation. CDP870 was used at concentrations ranging from 48.45 to 5001µg/ml. Approximately 2 hours prior to harvest, colchicine (1 µg/ml) was added to the cultures to arrest metaphase. The cells were fixed in methanol/acetic acid before slides were prepared. A repeat assay was conducted under the same conditions.

Analysis: Slides from selected treatments and from solvent and positive controls were coded using randomly generated letters. One hundred metaphases from each code were analyzed for chromosome aberrations. Only cells with 44-46 chromosomes were considered acceptable for analysis of structural aberrations. The aberrant cells in each culture were categorized as: (1) cells with structural aberrations including gaps, (2) cells with structural aberrations excluding gaps, and (3) polyploidy, endoreduplicated or hyperdiploid cells.

Criteria for Positive Results: The results were considered positive if the proportion of cells with structural aberrations at one or more concentrations exceeded the normal range in both replicate cultures, and a statistically significant increase in the proportion of cells with structural aberrations occurred at these concentrations.

RESULTS

Study Validity: The study was considered valid if the binomial dispersion test demonstrated acceptable heterogeneity between replicate cultures, the proportion of cells with structural aberrations (excluding gaps) fell within the historical control range, at least 160 of an intended 200 cells were analyzable at each dose level, and the positive controls induced statistically significant increases in the number of cells with structural aberrations. Based on the above mentioned criteria, the study was valid.

Study Outcome: Incubates containing CDP870 at 2823, 3751 and 5001 µg/ml concentrations were selected for analysis. There were 41% and 7% mitotic inhibitions at the highest concentration in the absence and presence of metabolic activation, respectively. Treatment of cultures with CDP870 in the absence and presence of S9 mix resulted in frequencies of cells with structural aberrations that were similar to and not significantly different from that of solvent controls, and these values fell within the historical control values from the conducting laboratory. The number of cells with chromosomal aberrations in the absence and presence of metabolic activation with 3 and 20 hours treatment are shown in the Tables below.

TABLE 1
3 hour treatment -S-9, 17 hour recovery (3+17), Experiment 1
Donor sex: Female

Treatment (μg/mL)	Replicate	Cells scored	Cells with aberrations including gaps	Cells with aberrations excluding gaps	Mitotic index (mean)
Solvent	A	100	1	1	7.3
	B	100	0	0	7.3
	Totals	200	1	1	(7.3)
2450	A	100	1	0	7.1
	B	100	2	1	7.3
	Totals	200	3	1	(7.2)
3501	A	100	1	1	5.8
	B	100	0	0	7.7
	Totals	200	1	1	(6.8)
5001	A	100	0	0	6.1
	B	100	2	2	5.9
	Totals	200	2	2	(6.0)
NQO, 2.5	A	100	14	13	
	B	100	22	18	
	Totals	200	36	31*	

Binomial Dispersion Test $\chi^2 = 5.04$, not significant

* Statistical significance $p \leq 0.001$

APPEARS THIS WAY
ON ORIGINAL

TABLE 2
3 hour treatment +S-9, 17 hour recovery (3+17), Experiment 1
Donor sex: Female

Treatment (μg/mL)	Replicate	Cells scored	Cells with aberrations including gaps	Cells with aberrations excluding gaps	Mitotic index (mean)
Solvent	A	100	2	1	9.1
	B	100	1	1	10.2
	Totals	200	3	2	(9.7)
2450	A	100	0	0	13.8
	B	100	0	0	12.8
	Totals	200	0	0	(13.3)
3501	A	100	1	1	11.1
	B	100	2	1	14.0
	Totals	200	3	2	(12.6)
5001	A	100	0	0	13.9
	B	100	0	0	11.3
	Totals	200	0	0	(12.6)
CPA, 12.5	A	100	28	27	
	B	100	24	22	
	Totals	200	52	49*	

Binomial Dispersion Test $\chi^2 = 0.00$, not significant

* Statistical significance $p \leq 0.001$

APPEARS THIS WAY
ON ORIGINAL

TABLE 3
20 hour treatment -S-9, 0 hour recovery (20+0), Experiment 2
Donor sex: Female

Treatment (μg/mL)	Replicate	Cells scored	Cells with aberrations including gaps	Cells with aberrations excluding gaps	Mitotic index (mean)
Solvent	A	100	1	0	5.8
	B	100	3	2	5.4
	Totals	200	4	2	(5.6)
2813	A	100	0	0	5.4
	B	100	0	0	4.2
	Totals	200	0	0	(4.8)
3751	A	100	0	0	4.8
	B	100	0	0	5.4
	Totals	200	0	0	(5.1)
5001	A	100	0	0	4.2
	B	100	1	0	2.8
	Totals	200	1	0	(3.5)
NQO, 2.5	A	100	35	33	
	B	100	32	30	
	Totals	200	67	63*	

Binomial Dispersion Test $\chi^2 = 2.02$, not significant

* Statistical significance $p \leq 0.001$

APPEARS THIS WAY
ON ORIGINAL

TABLE 4
3 hour treatment +S-9, 17 hour recovery (3+17), Experiment 2
Donor sex: Female

Treatment (μg/mL)	Replicate	Cells scored	Cells with aberrations including gaps	Cells with aberrations excluding gaps	Mitotic index (mean)
Solvent	A	100	4	2	8.8
	B	100	0	0	9.6
	Totals	200	4	2	(9.2)
2813	A	100	1	0	8.4
	B	100	1	0	9.1
	Totals	200	2	0	(8.8)
3751	A	100	0	0	9.1
	B	100	1	0	10.3
	Totals	200	1	0	(9.7)
5001	A	100	0	0	9.3
	B	100	0	0	8.2
	Totals	200	0	0	(8.8)
CPA, 45	A	100	35	33	
	B	80	27	22	
	Totals	180	62	55*	

Binomial Dispersion Test $\chi^1 = 2.02$, not significant

* Statistical significance $p \leq 0.001$

Thus, CDP870 was not mutagenic in the human peripheral blood lymphocytes chromosomal aberration assay, under the conditions of the experiment.

SUMMARY: CDP870 was not positive in the human peripheral blood lymphocytes chromosomal aberration assay in the absence or presence of metabolic activation.

APPEARS THIS WAY
ON ORIGINAL

Study Title: CDP870- Induction of Micronuclei in the Bone Marrow of Treated Mice

Sponsor's ID # Study Report No. 40000982

Descriptive Title: *In vivo* bone marrow micronucleus test in mice.

Conducting Laboratory: [redacted]

Date of Study Initiation/completion: March 30, 1998/ March 24, 1999

GLP Compliance: Yes

Drug Lot Number: CDP870, Lot no. 97LM202P

Study Endpoint: Frequency of micronucleated immature (polychromatic) erythrocytes.

METHODOLOGY:

Strains/Species/Cell line: CD-1 mice

Dose Selection Criteria: The highest dose administered in the range-finding study was 416.4 mg/kg. This was the highest dose that was possible to administer from the formulation supplied (based on a dosing volume of 20 ml/kg). In the range-finding study, the animals were dosed daily for two consecutiv days. The animals were observed for 2 days following the second administration for clinical signs and body weight changes. No clinical signs were observed in any animals. In the main study, CDP870 was administered at doses of 104.1, 208.2 and 416.4 mg/kg.

Test Agent Stability Considerations: The sponsor's stated concentration of the test article was 19.1 mg/ml in acetate buffer. Upon analysis of the test article, the concentration was found to be 20.82 mg/ml.

Metabolic Activation System: N/A.

Controls: Acetate buffer (pH 5.5) was used as a negative control. Cyclophosphamide, dissolved in physiological saline, was used a positive control.

Exposure conditions: The animals (8 animals/group) were administered CDP870 by a bolus i.v. injection at doses of 104.1, 208.2 and 416.4 mg/kg once daily for two consecutive days. The positive control administered as a single dose of 40 mg/kg on the second day of dosing. Twenty-four hours after the second administration, the animals were sacrificed and bone marrow samples collected from the femurs. Slides were prepared and fixed in methanol before staining with Giemsa stain.

Analysis: The number of micronucleated erythrocytes, polychromatic erythrocytes (PCE) and polychromatic and normochromatic erythrocytes (NCE) were counted. The proportions of polychromatic erythrocytes (PCE) and normochromatic erythrocytes (NCE) were determined by counting at least 2000 PCEs. The ratio of PCE/NCE for each animal and the mean for each group was calculated. The

individual and group mean frequency of micronucleated PCE/1000 cells was also determined. The numbers of micronucleated PCE in each treatment group were then compared with the numbers in vehicle control groups by using a 2X2 contingency table to determine $\chi 2$.

Criteria for Positive Results: The results were considered positive when there were significant and dose-dependent increases in the MNPCE in the treatment groups as compared to the negative control.

RESULTS

Study Validity: The study was considered valid when the frequencies of micronucleated PCEs in the negative control group fell within the historical control range from the conducting laboratory, at least seven animals from each group were analyzable, and the positive control induced a statistically significant increase in the frequency of micronucleated PCEs. Based on these criteria, the study was valid.

Study Outcome: There was no significant increase in the frequency of MNPCE in any treatment groups as compared with the negative control group. Groups of mice treated with CDP870 exhibited PCE/NCE ratios that were similar to the vehicle control group. The group mean frequencies of micronucleated PCE/1000 cells are shown in the Table below.

Data for CDP 870

Treatment group (mg/kg/day)	Kill time (hours)	Sex	Mean ratio PCE/NCE	Group mean frequency of micronucleated PCE (per 1000)
				per treatment group (±sd)
Vehicle control	24	♂	0.88	0.06 ± 0.15
Untreated control	24	♂	0.83	0.17 ± 0.21
104.1	24	♂	0.88	0.00 ± 0
208.2	24	♂	0.77	0.06 ± 0.15
416.4	24	♂	0.75	0.00 ± 0
CPA, 40+	24	♂	0.56	5.46 ± 7.2

+ Administered as a single dose
sd Standard deviation

Thus, CDP870 was not positive in the mouse bone marrow micronucleus assay at i.v. doses up to 416.4 mg/kg/day under the experimental conditions.

Summary:

The genotoxic potential for CDP870 was assessed in the bacterial reverse mutation assay (Ames assay), the human peripheral blood lymphocyte chromosomal aberration assay and the mouse bone marrow micronucleus assay. CDP870 had no genotoxic potential in any of these assays.

2.6.6.5 Carcinogenicity

No carcinogenicity studies were conducted with CDP870.

2.6.6.6 Reproductive and developmental toxicology

Study title: Anti-Rat TNFα Monoclonal Antibody Equivalent to CDP870- Fertility Study by the Intravenous Route in the Rat (Segment I)

Study Report Nos. 40001308 and 40001216

Conducting laboratory (and location if not the Sponsor): [redacted]

Dates of study initiation & completion: January 11, 2002 & June 24, 2003

GLP compliance: Yes

QA Report Yes (X) No ( )

Drug, Lot #, radiolabel (if applicable), and % purity: [redacted] : anti-murine TNF monoclonal antibody equivalent to CDP870 (cTN3 PF); Batch # 10014367/53.

Protocol reviewed by the Division? No

METHODS:

Species: Sprague-Dawley rats Ico: OFA.SD (IOPS Caw)

Doses employed and Route of Administration: Three groups of rats were intravenously administered (infused over 2 minutes) 0 (acetate buffer), 20 or 100 mg/kg doses of cTN3 PF twice weekly. Female animals were dosed twice weekly for 2 weeks before pairing, during the mating period and on days 1 and

4 of gestation. Males were dosed twice weekly for 4 weeks before, throughout the second 2-week mating period and for further four weeks. Satellite females were dosed twice weekly for 4 weeks, and satellite males were dosed twice weekly for 10 weeks.

Study Design: The test article was administered intravenously (2 ml/kg) into a tail vein at doses of 20 and 100 mg/kg. Female animals were dosed twice weekly for 2 weeks before pairing, during the mating period and on days 1 and 4 of gestation. Males were dosed twice weekly for 4 weeks before, throughout the second 2-week mating period and for further four weeks. The sponsor stated that due to technical problems, some animals were treated subcutaneously for the following occasions: male #98 (control) on day 10; Male #109 (50 mg/kg) and #132 (100 mg/kg) on day 14; male #87 (control) on day 21, and male #130 and #141 (100 mg/kg) on day 18.

Number of animals/sex/dosing group: 25 animals/sex/group in the main study, and 3 animals/sex/group in the satellite groups.

Parameters and endpoints evaluated: The animals were observed once a day for general signs and mortalities. The body weights of the males were measured twice a week. The body weights of the females were measured twice a week during the premating and mating periods and on days 0, 4, 8 and 13 of gestation. Food consumption of treated females was measured weekly during the pre-mating and mating periods, and on days 0, 4, 8 and 13 of gestation. Food consumption of males was measured weekly. Mating was confirmed by the presence of sperms in the vaginal smears. Male animals were sacrificed on the day following the last dosing and necropsies performed. The testes and epididymides were weighed and used for sperm analysis. The female animals were sacrificed on Day 13 of pregnancy and necropsies performed. The number of corpora lutea and implantation were counted and the ovaries and the uteri were fixed in buffered formalin. The following organs were subjected to histopathological examinations: vagina, cervix, uterus, ovary, pituitary gland, seminal vesicles, prostate, coagulating gland, right testes, right epididymis and left caput epididymis. Blood samples were collected from all main study animals at terminal necropsy for determination of plasma concentrations of cTN3 PF and antibodies to cTN3 PF. Blood samples from the satellite animals were collected on days 0 (males and females) and on one day during week 4 (females) and 6 and 10 (males) before and 15 minutes after treatment.

RESULTS:

Clinical signs: No treatment-related clinical signs were observed in males and females from any group.

Estrous cycle: There was no adverse effect of treatment with the TNF-α antibody on the estrous cycle of animals from any group

Mortality: There were no unscheduled deaths in any groups.

Body weight and food consumption: The initial mean body weight of the control males and females were 319.9±10.4 and 240.9±13.0 g respectively. The final body weights of the males and females were 509.2±26.2 and 323.9±21.2 g respectively. No treatment-related effects on the body weights were observed in any group. Treatment with the test substance had no adverse effect on food consumption of the animals.

Toxicokinetics in parental animals: Mean plasma concentrations of cTN3 PF of male and female animals increased with increasing dose, but was not always dose-proportional. The plasma concentrations of the male animals were higher than that for females. The differences may be due to different dosing schedules for males and females. The plasma concentrations of the main study animals are summarized in the Table below.

Gender	cTN3 PF (µg/mL)		
	0 mg/kg	20 mg/kg	100 mg/kg
Male	0.02	266.2	771.4
Female	0.02	26.9	103.4

Antibodies were detectable in one rat (female #49, receiving 20 mg/kg; 0.4 units/ml). The antibody concentrations in other samples were below the lower limit of quantification (— units/ml).

The plasma concentrations of cTN3 PF in male and female satellite group animals appeared to be similar. The plasma concentrations of the satellite animals are summarized in the Table below.

Time of sample collection	Males		Females	
Week	20 mg/kg	100 mg/kg	20 mg/kg	100 mg/kg
0 pre-injection	0.02	0.02	0.02	0.02
0 post-injection	351.4	1588.3	358.7	1332.8
4 pre-injection	-	-	139.3	556.2
4 post-injection	-	-	497.3	2190.7
6 pre-injection	128.3	624.1	-	-
6 post-injection	455.9	2160.8	-	-
10 pre-injection	139.5	597.2	-	-
10 post-injection	512.5	1468.4	-	-

Antibodies to cTN3 PF were not detected in samples from the satellite animals.

Fertility in Males: There were no treatment-related effects on the mating performance of the animals. There were no differences in sperm counts and sperm motility between the control and the treatment groups. There were no differences in the histopathological findings between the control and the treatment groups.

Fertility and Early Embryonic Development in Females: Treatment with cTN3 PF had no effect on the fertility of the animals. All inseminated females became pregnant with the exception of one control female. The number of corpora lutea and uterine implantations and the corresponding pre-implantation loss were comparable in all groups. There were no adverse effects of the treatment on embryo survival in any group. The mean live litter size was comparable in all groups for the treated females and the untreated females paired with the treated males. The effect of cTN3 PF on mating performance and fertility of female rats are summarized in the Table below.

MATING PERFORMANCE AND FERTILITY - SUMMARY
Females

	Group 1 Control 0 mg/kg	Group 2 Low dose 20 mg/kg	Group 3 High dose 100 mg/kg
NUMBER OF FEMALES:			
Paired	25	25	25
Inseminated	25	24	24
Pregnant	24	24	24
PRE-COITAL INTERVAL - DAYS			
MEAN	2.40	2.71	2.50
S.D.	1.12	1.27	1.02
N	24	24	24
COPULATION INDEX (%)	100	96	96
FEMALE FERTILITY INDEX (%)	96	100	100

The effects of cTN3 PF on the number of corpora lutea, implantations, viable fetuses and pre- and post-implantation losses are shown in the Table below.

SUMMARY OF CAESAREAN SECTION DATA

	Group 1 Control 0 mg/kg	Group 2 Low dose 20 mg/kg	Group 3 High dose 100 mg/kg
Number of dams	24	24	24
Corpora Lutea			
per group	395	376	388
per dam: mean	16 d	16	16
st. dev.	2	2	2
Implantations			
per group	378 f	360	372
% of Corpora lutea	95.7	95.7	95.9
per dam: mean	16 d	15	16
st. dev.	2	2	2
Viable Embryos			
per group	357	339	359
per dam: mean	15 d	14	15
st. dev.	2	2	2
% of Implantations			
per group	94.4 f	94.2	96.5
per dam: mean	95 d	94	96
st. dev.	5	7	5
Postimplantation Loss			
total per group	21 f	21	13
% of implantations	5.6	5.8	3.5
per dam: mean	1 k	1	1
st. dev.	1	1	1
No. of dams affected	15	13	9
Preimplantation Loss			
per group	17 f	16	16
% of Corpora lutea	4.3	4.3	4.1
per dam: mean	1 k	1	1
st. dev.	1	1	1
No. of dams affected	15	13	13

Statistical key: d=Anova/Dunnett test f=Chi2/Fisher Exact test k=Kruskal-Wallis/Dunn test

In the Segment I fertility and early embryonic development study in rats, male and female animals were treated with 20 and 100 mg/kg i.v. doses of cTN3 PF (anti-rat TNFα monoclonal antibody equivalent to CDP870), administered twice weekly. Treatment of male and female rats with cTN3 PF had no effects on gonadal function, mating behavior and reproductive performance of the animals. The number of implantation sites and live embryos were similar in all groups, including the control. Thus, cTN3 PF had no effects on the fertility and early embryonic development in rats.

Study Title: Anti-Rat TNFα Monoclonal Antibody Equivalent to CDP870- Embryo toxicity Study by the Intravenous Route in the Rat (Segment II)

Study Report No # 40001271 and # 40001119.

Conducting laboratory and location:

Dates of study initiation and completion: April 4, 2001 and October 2, 2002.

GLP compliance: Yes

QA Report Yes (X) No ( )

Drug, Lot #, radiolabel, and % purity: Mouse/hamster anti-murine monoclonal antibody equivalent to CDP870 — TN3 pegylated Fab (cTN3 PF); Batch no. 10014166/75; purity, 100%.

Methods:

Species/strain: —: CD (SD)IGS BR VAF/Plus® rats.

Doses employed: Two groups of 25 mated rats were intravenously administered 20 (group 3) and 100 (group 5) mg/kg doses of anti-rat TNFα monoclonal antibody equivalent to CDP870 on days 1 and 4 of gestation. Two other groups were treated at the same dose levels on days 6, 9, 13 and 16 of gestation (groups 4 and 6). Two other groups of rats were treated with the vehicle (60 mM sodium acetate buffer) only on the same days (groups 1 and 2).

Route of administration: Intravenous.

Study design: Two groups of pregnant rats received 20 and 100 mg/kg i.v. doses of anti-rat TNFα monoclonal antibody equivalent to CDP870 on days 1 and 4 of gestation. Two other groups received same doses of the antibody on gestation days 6, 9, 13 and 16. Two control groups were treated with the vehicle on the same days as the test substance. The design of the study is summarized in the Table below.

Group/ treatment	Nominal dose level (mg/kg)	Dose volume (ml/kg)	Nominal dose concentration (mg/ml)	Dosing days (1)
1. Control 1	0	2	0	1 and 4
2. Control 2	0	2	0	6, 9, 13 and 16
3. Low dose 1	20	2	10	1 and 4
4. Low dose 2	20	2	10	6, 9, 13 and 16
5. High dose 1	100	2	50	1 and 4
6. High dose 2	100	2	50	6, 9, 13 and 16

(1) Days of gestation

Number/sex/group: 25 female animals/group.

Parameters and endpoints evaluated: The animals were observed once daily for clinical signs and mortality. The body weights were recorded on gestation days 0, 6, 11, 13, 15, 18 and 20. Food consumption was recorded on gestation days 0 to 6, 6 to 11, 11 to 13, 13 to 15, 15 to 18 and 18 to 20. On day 20 of gestation, all surviving dams were sacrificed and their uterine contents, including the placentae were examined. The females were examined macroscopically and live fetuses were weighed, sexed and examined for visceral and skeletal abnormalities. Half of the fetuses were examined internally prior to processing for skeletal examination. The remaining fetuses were fixed for visceral examination by the modified Wilson-Barrow technique.

Toxicokinetics: Blood samples for determination plasma levels of cTN3 PF were collected on days 5, 6, 7, 9, 11, 13, 16, 18 and 20 of gestation. The animals were sampled immediately after treatment on days 6, 9, 13 and 16 of gestation. All main group animals were sampled at terminal necropsy on day 20 of gestation. Blood samples were also collected from fetuses from the satellite group (group 6) animals.

Results:

In-life observations:

Mortality: There were no mortalities in any group.

Clinical signs: No treatment related clinical signs were observed in any group throughout the study period.

Body weight: The mean body weights of the control 1 animals before initiation dosing and at the end of the dosing period were 234.4±13.4 and 400.7±27.1 g, respectively. Treatment of the animals with cTN3 PF had no adverse effects on the mean body weights.

Food consumption: No treatment-related changes in food consumption were observed in any group.

Terminal and macroscopic evaluations:

There were 4 non-pregnant females in each of the low dose 2 and high dose 2 groups compared with one or two in the control groups. One of the females from the low dose 2 group had 10 corpora lutea with no visible implantation sites. The numbers of corpora lutea and implantation sites were comparable in all groups. One female in each of the control 2 (#50) and low dose 1 (#72) groups had no viable fetuses. The mean live litter size was slightly reduced in the control 2 group compared with the other groups. Mean fetal weight and fetal sex ratio were comparable in all groups. Cesarean section data for different groups are summarized in the Table below.

SUMMARY OF CAESAREAN SECTION DATA

		Group 1 Control 1 0 mg/kg	Group 2 Control 2 0 mg/kg	Group 3 Low dose 1 20 mg/kg	Group 4 Low dose 2 20 mg/kg	Group 5 High dose 1 100 mg/kg	Group 6 High dose 2 100 mg/kg
Pregnant	N	23	24	25	21	23	21
Dams with no Viable Fetuses	N	0	1	1	0	0	0
Dams with Viable Fetuses	N	23	23	24	21	23	21
Corpora Lutea	TOTAL	378	359	383	343	375	329
No. per animal	MEAN	16.4 k	15.0	15.3	16.3	16.3	15.7
	S.D.	3.1	4.0	3.9	3.7	1.9	2.4
Implantation Sites	TOTAL	321	278	324	271	309	279
No. per animal	MEAN	14.0 d	11.6	13.0	12.9	13.4	13.3
	S.D.	3.5	4.2	3.7	2.8	3.0	3.3
Preimplantation Loss	TOTAL	57	81	59	72	66	50
No. per animal	MEAN	2.5 d	3.4	2.4	3.4	2.9	2.4
	S.D.	2.7	3.0	2.6	4.4	2.7	3.9
% per animal	MEAN%	14.8 k	23.4	16.3	18.4	17.5	14.2
	S.D.	17.4	20.2	18.1	20.4	16.8	21.6
Live Fetuses	TOTAL	289	248	290	251	285	252
No. per animal	MEAN	12.6 d	10.3	11.6	12.0	12.4	12.0
	S.D.	3.6	4.4	3.7	2.9	3.1	3.1
Males	TOTAL	135	123	158	110	149	112
	MEAN%	46.1 k	49.0	56.2	42.7	52.4	44.3
	S.D.	16.7	15.7	16.4	16.0	17.0	18.0
Females	TOTAL	154	125	132	141	136	140
	MEAN%	53.9 k	51.0	43.8	57.3	47.6	55.7
	S.D.	16.7	15.7	16.4	16.0	17.0	18.0

Statistical key: d=Anova/Dunnett test k=Kruskal-Wallis/Dunn test

SUMMARY OF CAESAREAN SECTION DATA

		Group 1 Control 1 0 mg/kg	Group 2 Control 2 0 mg/kg	Group 3 Low dose 1 20 mg/kg	Group 4 Low dose 2 20 mg/kg	Group 5 High dose 1 100 mg/kg	Group 6 High dose 2 100 mg/kg
Postimplantation Loss	TOTAL	32	30	34	20	24	27
No. per animal	MEAN	1.4 d	1.3	1.4	1.0	1.0	1.3
	S.D.	0.9	1.2	1.0	0.8	1.0	1.0
% implants per animal	MEAN%	10.5 k	15.4	13.2	8.1	8.3	9.5
	S.D.	8.5	22.2	19.5	7.5	7.5	6.8
Dead Fetuses	TOTAL	0	0	0	0	0	0
No. per animal	MEAN	0.0 k	0.0	0.0	0.0	0.0	0.0
	S.D.	0.0	0.0	0.0	0.0	0.0	0.0
% of implants per animal	MEAN%	0.0 k	0.0	0.0	0.0	0.0	0.0
	S.D.	0.0	0.0	0.0	0.0	0.0	0.0
Resorptions: Early	TOTAL	32	30	34	19	23	26
No. per animal	MEAN	1.4 k	1.3	1.4	0.9	1.0	1.2
	S.D.	0.9	1.2	1.0	0.8	1.0	0.9
% of implants per animal	MEAN%	10.5 k	15.4	13.2	7.8	8.1	9.2
	S.D.	8.5	22.2	19.5	7.5	7.5	6.5
Resorptions: Late	TOTAL	0	0	0	1	1	1
No. per animal	MEAN	0.0 k	0.0	0.0	0.0	0.0	0.0
	S.D.	0.0	0.0	0.0	0.2	0.2	0.2
% of implants per animal	MEAN%	0.0 k	0.0	0.0	0.3	0.3	0.3
	S.D.	0.0	0.0	0.0	1.4	1.2	1.4

Statistical key: d=Anova/Dunnett test k=Kruskal-Wallis/Dunn test

A total of 289, 248, 290, 251, 285 and 252 fetuses were available for examination in groups 1, 2, 3, 4, 5 and 6, respectively. There was single malformed fetus in each of groups 1, 2, 3 and 6. The fetus from control 1 female (#25) had acaudia, the fetus from control 2 female (#30) had anasarca, paw/limb defects (bradydactyly, flexed right forelimb and malrotated hindlimbs) and an absent genital tubercle. The fetus from a low dose female (#54) had multiple defects including open eyes, a protruding tongue, excencephaly and spina bifida, and the fetus from a high dose 2 female (#146) had a proboscis with an associated eye defect.

Visceral examination revealed four further malformed fetuses, three of which were from the same low dose 2 female (#83). Each of these fetuses had a small right and enlarged left kidney, respectively, and in each case one or both of the kidneys were displaced caudally. The other fetus, also from low dose 2 group (female #93), had a cleft palate and malformed kidneys (both kidneys were small, misshapen and displaced caudally). However, these effects were not dose-related and observed in the control and low dose groups. Fetal malformations in different groups are summarized in the Table below.

Group	Female number	Fetus number(s)	Malformation(s)
1 Control 1	25	5	Acaudia, malformed vertebral column
		13	Malformed vertebral column
2 Control 2	30	3	Anasarca, brachydactyly, absent genital tubercle, small mandible
3 Low dose 1	54	4	Open eyes, exencephaly, spina bifida, cleft palate, malformed kidneys
4 Low dose 2	83	6	Malformed kidneys
		10	Malformed kidneys
		12	Malformed kidneys
	93	10	Cleft palate
6 High dose 2	146	1	Proboscis, malpositioned eyes,

Skeletal examination revealed one malformed fetus from a control 1 female (#25). The fetus had a gross disruption of the vertebral column from the first thoracic vertebra with little evidence of ossification thereafter.

Toxicokinetics: Following intravenous administration to pregnant rats, the mean plasma concentrations of cTN3 PF increased with increasing doses. The mean plasma concentrations for groups 3 and 5 (20 and 100 mg/kg, respectively) dosed on gestation days 1 and 4 were 0.235 and 1.384 µg/ml, respectively, on gestation day 20. Groups 4 and 6, which received 20 and 100 mg/kg on 4 occasions, had mean plasma concentrations of 20.605 and 75.282 µg/ml, respectively, on gestation day 20. Between infusions, the mean plasma levels remained high during the study period (584.354 µg/ml on gestation day 7 and 464.584 µg/ml on gestation day 11). The mean plasma concentrations of the fetuses from 3 satellite dams were 0.251µg/ml, less than 0.3% of the

maternal level on gestation day 20. The mean plasma concentrations of animals from different groups on gestation day 20 are shown in the Table below.

Summary Statistics of cTN3 PF Concentrations (μg/mL) at Day 20 of Gestation

	Dose (mg/kg)		
	0	20	100
Group (Dosing Days)	1(G1, G4)	3(G1, G4)	5(G1, G4)
N	23	25	23
Mean	0.020	0.235	1.384
95% CI	(0.020, 0.020)	(0.203, 0.270)	(1.157, 1.656)
Group (Dosing Days)	2(G6, G9, G13, G16)	4(G6, G9, G13, G16)	6(G6, G9, G13, G16)
N	24	21	21
Mean	0.020	22.605	75.282
95% CI	(0.020, 0.020)	(19.312, 26.460)	(63.384, 89.412)

Note: The geometric mean and its associated 95% confidence interval are presented.
The limit of quantification is — μg/mL, values below the limit of quantification are set to half the limit of quantification for the calculation of summary statistics.
Rats 4, 7, 35, 81, 91, 92, 96, 108, 118, 127, 132, 136 and 149 are excluded from summary statistics because they were not pregnant.
VERSION: 1

Summary:

In the segment II teratogenicity study with anti-rat TNFα monoclonal antibody equivalent to CDP870 (cTN3 PF) in rats, the test agent was administered intravenously at 20 and 100 mg/kg doses on gestation days 1 and 4, or gestation days 6, 9, 13 and 16. cTN3 PF was not teratogenic in rats at the doses examined.

Study Title: Anti-Rat TNFα Monoclonal Antibody (cTN3 PF) Equivalent to CDP870- Pre- and Post-natal Development Study by the Intravenous Route in the Rat (Segment III).

Study Report No # 40001534

Conducting laboratory and location: [redacted]

Dates of study initiation and completion: September 27, 2004 and November 21, 2005.

GLP compliance: Yes

QA Report Yes (X) No ( )

Drug, Lot #, radiolabel, and % purity: [redacted] monoclonal antibody equivalent to CDP870/cTN3 PEGylated Fab (cTN3 PF); Batch no. 10016740/05; purity, 100%.

Methods:

Species/strain: Sprague-Dawley rats- [redacted] OFA.SD (IOPS Caw).

Doses employed: 30 and 100 mg/kg, twice weekly (on days 6, 9, 13, 16 and 20 of gestation and days 2, 6, 9, 13, 16 and 20 of lactation).

Route of administration: Intravenous

Study design: Two groups of 25 female time-mated rats were administered cTNF PF at dose levels of 30 and 100 mg/kg twice weekly from day 6 of gestation through weaning (day 20 of lactation). Another group of 25 rats received the vehicle only (50 mM sodium acetate buffer/125 mM NaCl) over the same period, and served as a control. The study design is summarized in the Table below.

Group/ Treatment	Dose level (mg/kg/adminstration)	Dose volume (mL/kg)	Dose concentration (mg/mL)
1. Control	0	2.38	0
2. Low dose	30	2.38	12.6
3. High dose	100	2.38	42.1

Only females of the F0 generation were treated.

Number/sex/group: 25 animals/group.

Parameters and endpoints evaluated: The animals were observed twice daily for clinical signs and mortality. The body weights of the F0 animals were recorded on days 0, 6, 11, 15, 18 and 20 of gestation and on days 1, 4, 7, 10, 14, 17 and 21 of lactation. Food consumption of individual animal was recorded on days 0 to 6, 6 to 11, 11 to 15, 15 to 18 and 18 to 20 of gestation, and on days 1 to 4, 4 to 7, 7 to 10, 10 to 14 and 14 to 21 of

lactation. For each F0 female, the following data were recorded: date of mating, date of parturition, duration of gestation, abnormalities of delivery, nesting or nursing behavior and number of implantation sites. The weights of the following organs of the FO dams were recorded: adrenal glands, brain, lungs, heart, thymus, kidneys, liver, ovaries, prostate, adrenals, pituitary gland, spleen, testes, thyroid glands. The following tissues were examined histologically: thymus, spleen, lymph nodes (mandibular, mesenteric), peyer's patches and all gross lesions.

For each F1 litter, the following data were recorded: number of male and female pups born, external abnormalities, number, weight and sex of pups on days 1, 4, 7, 10, 14, 17 and 21 post-partum, physical development of the offspring (duration of pinna unfolding, incisor eruption and eye opening), behavior and functional tests in all pups (surface righting reflex on day 8 post-partum, gripping reflex on day 17, pupil reflex and auditory reflex on day 21) and external necropsy findings of dead pups.

On day 4 post-partum, the size of each litter was adjusted (culling) to yield 4 males and 4 females per litter and divided into two subgroups. After weaning of the F1 pups, at least 2 male and 2 female pups per litter, where possible, were selected for the post-weaning behavioral tests and mating (25 males and 25 females per group). The following post-weaning development tests were performed: Females were examined from day 28 post-partum for vaginal opening; males were examined from day 38 post-partum to detect the day of balano preputial skinfold cleavage. In addition, water maze (5 to 6 weeks of age) and open field (7 weeks of age) were performed on selected offspring of the first sub-group.

The F1 animals (25 males and 25 females) were mated at approximately 12 weeks of age for up to 12 days. F1 males were examined macroscopically. F1 females were sacrificed on day 13 post-coitum and examined macroscopically for pregnancy status, number of corpora lutea, and numbers and types of uterine implantations.

Toxicokinetics: A terminal blood sample was collected from F0 dams after collection of milk on day 22 of lactation for determination of cTN3 PF concentrations and antibodies to cTN3. Terminal blood samples were also collected at the time of necropsy from the 2nd sub-group of F1 adult animals (approximately 9 weeks of age).

Where possible, milk samples were taken from each lactating F0 female prior to termination (day 22 of lactation) for determination of cTN3 PF concentrations and antibodies to cTN3.

Results:

Mortality: There were no treatment-related mortalities of F0 dams in any group. One control group dam was found dead on day 22 of gestation.

Clinical signs: No treatment related clinical signs were observed in any group.

Body weight: The mean body weights of the control dams were 222.7±8.2 and 395.4±21.4 g before initiation of dosing and day 20 of gestation, respectively. Treatment of F0 pregnant

animals with intravenous cTN3 PF had no adverse effects on the body weights of the animals.

Food consumption: Mean food consumptions of the control animals were 23.9±2.2 and 27.8±2.6 g/animal/day on days 1-6 and 6-20, respectively. The mean food consumptions were comparable in all groups during the treatment period.

In-life observations: The pregnancy rates were similar in all groups. The duration of gestation was comparable in the control and treatment group animals. Of the surviving females, a total of 24, 22 and 23 successfully reared their young to weaning in groups 1, 2 and 3, respectively. Delivery and litter data for different groups are summarized in the Table below.

SUMMARY OF DELIVERY AND LITTER DATA

		Group 1 Control 0 mg/kg		Group 2 Low dose 30 mg/kg	Group 3 High dose 100 mg/kg
Females on Study	N	25		25	25
Females Mated	N	25	f	25	25
Mating Index	%	100.0		100.0	100.0
Females Pregnant	N	25	f	23	24
Female Fertility Index	%	100.0		92.0	96.0
Females with Liveborn	N	24	f	22	23
Gestation Index	%	96.0		95.7	95.8
Females Completing Delivery	N	24	f	23	23
	%	96.0		92.0	92.0
with Stillborn Pups	N	4	f	4	6
	%	16.7		17.4	26.1
with all Stillborn	N	0	f	1	0
	%	0.0		4.3	0.0
Litters with Liveborn, but no Pups Alive					
day 4	N	0	f	0	0
	%	0.0		0.0	0.0
day 21	N	0	f	0	0
	%	0.0		0.0	0.0
Duration of Gestation	MEAN	21.9	d	22.0	22.0
	S.D.	0.3		0.5	0.3
	N	24		23	23

Statistical key: d=Anova/Dunnett test f=Chi2/Fisher Exact test

SUMMARY OF DELIVERY AND LITTER DATA

		Group 1 Control 0 mg/kg		Group 2 Low dose 30 mg/kg	Group 3 High dose 100 mg/kg
Litters with Liveborn Pups	N	24		22	23
Pups Delivered (total)	N	293		265	283
	MEAN	12.2	d	11.5	12.3
	S.D.	1.9		3.4	3.5
Liveborn	N	289	f	255	274
Live Birth Index	%	98.6		96.2	96.8
Stillborn	N	4	f	10	9
	%	1.4		3.8	3.2
Culled day 4		93		86	100
Liveborn, not culled prior to day 21	N	196		169	174
Pups Dying, Missing, and/or Cannibalized					
day 0	N	0	f	0	0
	%	0.0		0.0	0.0
days 1-4	N	4	f	2	0
	%	1.4		0.8	0.0
days 5-21	N	2	f	2	1
	%	0.7		0.8	0.4
days 0-4	N	4	f	2	0
	%	1.4		0.8	0.0
days 0-21	N	6	f	4	1
	%	2.1		1.6	0.4
Pups Surviving 4 days	N	285	f	253	274
Viability Index	%	98.6		99.2	100.0
Pups Surviving 21 days	N	190	f	165	173
Lactation Index	%	99.0		98.8	99.4

Statistical key: d=Anova/Dunnett test f=Chi2/Fisher Exact test

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The mean body weights of pups on day 1 and day 4 are summarized in the Table below.

Age	Control	30 mg/kg	100 mg/kg
Day 1			
-males	8.1±0.7	8.0± 0.7	7.8 ± 0.7
-females	7.7± 0.7	7.8 ± 0.8	7.5 ± 0.7
Day 4			
-males	11.7 ± 1.1	11.5 ± 1.3	11.4 ± 1.4
-females	11.2 ± 1.1	11.0 ± 1.4	11.0 ± 1.3
Day 7			
-males	18.5 ± 1.5	18.2 ± 1.9	18.3 ± 1.8
-females	17.7 ± 1.5	17.5 ± 2.3	17.5 ± 1.5
Day 10			
-males	25.6 ± 1.7	25.8 ± 2.4	25.7 ± 2.3
-females	25.3 ± 1.9	24.6 ± 3.6	24.9 ± 2.1
Day 14			
-males	35.6 ± 1.9	36.3 ± 2.6	36.2 ± 3.2
-females	34.5 ± 2.3	34.3 ± 5.0	34.9 ± 3.0
Day 17			
-males	42.6 ± 2.4	42.8 ± 3.1	43.4 ± 3.6
-females	40.9 ± 2.8	40.6 ± 6.1	41.9 ± 3.4
Day 21			
-males	56.9 ± 3.4	56.8 ± 4.3	57.2 ± 4.9
-females	54.3 ± 4.5	53.9 ± 7.8	55.2 ± 4.9

Terminal and macroscopic evaluations:

Dams: There were no treatment-related gross abnormalities in any group. There were no adverse effects of treatment on embryo-fetal survival (pre-birth loss). One female from the 30 mg/kg group had total litter death at birth.

Offspring:

Treatment of the F0 dams during periods of gestation and lactation had no effects on embryo-fetal survival, body weight or physical and functional development (surface righting, gripping, auditory or pupil reflexes) of the lactating pups through weaning (as assessed by the age of sexual maturation, performance in the water maze and open field tests and immune function (as assessed by the lymphocyte subset counts and plaque

forming cell assay) or their mating performance and fertility. Reflex and physical development for pups from different groups are summarized in the Table below.

REFLEX AND PHYSICAL DEVELOPMENT - SUMMARY

		Group 1 0 mg/kg	Group 2 30 mg/kg	Group 3 100 mg/kg
PINNA UNFOLDING - % of pups positive:				
day 1	*post-partum*	0	5	1
day 2	*post-partum*	73	70	71
day 3	*post-partum*	99	97	98
day 4	*post-partum*	100	100	100
INCISOR ERUPTION - % of pups positive:				
day 7	*post-partum*	0	2	0
day 8	*post-partum*	5	10	9
day 9	*post-partum*	33	32	33
day 10	*post-partum*	65	67	64
day 11	*post-partum*	94	92	95
day 12	*post-partum*	98	99	97
day 13	*post-partum*	100	100	100
EYE OPENING - % of pups positive:				
day 12	*post-partum*	2	0	1
day 13	*post-partum*	6	9	9
day 14	*post-partum*	52	62	51
day 15	*post-partum*	89	95	95
day 16	*post-partum*	98	100	100
day 17	*post-partum*	100		

	Group 1 0 mg/kg	Group 2 30 mg/kg	Group 3 100 mg/kg
SURFACE RIGHTING REFLEX - day 8 *post-partum*			
- % of pups positive:	100	100	100
GRIPPING REFLEX - day 17 *post-partum*			
- % of pups positive:	100	100	100
PUPILLARY REFLEX - day 21 *post-partum*			
- % of pups positive:	100	99	100
AUDITORY REFLEX - day 21 *post-partum*			
- % of pups positive:	100	100	100

There were 25, 24 and 25 pregnant F1 females derived from F0 groups treated with 0, 30 and 100 mg/kg doses respectively. There was no influence of maternal treatment on early embryonic development of pregnant F1 females. The incidence of embryonic death and the resulting mean live litter size were comparable in all groups. Terminal investigations of the F1 animals did not show any organ weight or macroscopic changes for either sex. Mating performance and fertility index for F1 animals from different groups are summarized in the Table below.

MATING PERFORMANCE AND FERTILITY - SUMMARY

GROUP/ TREATMENT	1 0 mg/kg	2 30 mg/kg	3 100 mg/kg
NUMBER OF FEMALES:			
Paired	25	25	25
Inseminated	25	25	25
Pregnant	11[1]	24	25
PRE-COITAL INTERVAL - DAYS			
MEAN	2.72[2]	2.56	3.08
S.D.	1.10[2]	1.73	2.18
N	25[2]	25	25
COPULATION INDEX (%)	100	100	100
FERTILITY INDEX (%)	100[1]	96	100

[1]: given that raw data sheets for 14/25 group 1 females were lost, results include animals with confirmed caesarean section.

[2]: calculated using all animals with positive evidence of insemination.

Cesarean section data for F1 dams from different groups are summarized in the Table below.

SUMMARY OF CAESAREAN SECTION DATA

	Group 1 Control 0 mg/kg	Group 2 Low dose 30 mg/kg	Group 3 High dose 100 mg/kg
Number of dams	11	24	25
Corpora Lutea			
per group	180	416	437
per dam: mean	16 d	17	17
st. dev.	2	3	3
Implantations			
per group	171 f	404	408
% of Corpora Lutea	95.0	97.1	93.4
per dam: mean	16 d	17	16
st. dev.	2	3	3
Viable Embryos			
per group	164	383	387
per dam: mean	15 d	16	15
st. dev.	2	3	4
% of Implantations			
per group	95.9 f	94.8	97.0
per dam: mean	96 k	94	93
st. dev.	4	8	20
Postimplantation Loss			
total per group	7 f	21	21
% of implantations	4.1	5.2	5.1
per dam: mean	1 k	1	1
st. dev.	1	1	2
No. of dams affected	6	12	10
Preimplantation Loss			
per group	9 f	12	29
% of Corpora Lutea	5.0	2.9	6.6
per dam: mean	1 k	1	1
st. dev.	1	1	1
No. of dams affected	6	7	16

Statistical key: d=Anova/Dunnett test f=Chi2/Fisher Exact test k=Kruskal-Wallis/Dunn test

Histological examinations of the thymus, lymph nodes, peyer's patches and spleen of the F1 animals did not show any inter-group differences in development of the lymphoid organs.

Toxicokinetics: The plasma concentrations of the F0 dams increased with increasing dose. The mean plasma concentrations of 30 and 100 mg/kg groups on day 22 were 225.48 and 702.79 µg/ml, respectively. There were no detectable antibodies to cTN3 PF in the plasma of dams from any group. Limited transfer of cTN3 PF to the milk was observed. Milk cTN3 PF concentrations on day 22 of dams treated with 30 and 100 mg/kg doses were 17.94 and 69.16 µg/ml, respectively. There were no detectable cTN3 PF in plasma samples from F1 animals at either study day 21 or 9 weeks post-partum. The plasma and milk concentrations (µg/ml) of cTN3 PF in animals from different groups are summarized in the Table below.

	Control	30 mg/kg	100 mg/kg
F0 Animals			
-plasma	0.02 ±0.0	225.48±54.54	702.79±106.26
-milk	0.02± 0.0	17.94±3.56	69.26±7.42
F1 animals (males)			
-plasma (week 9)	0.02±0.0	0.02±0.0	0.02±0.0
F1 animals (females)			
-plasma (week 9)	0.02±0.0	0.02±0.0	0.02±0.0

In the segment III pre- and post- natal development study with cTN3 PF in rats, groups of pregnant females were intravenously administered the antibody at 0, 30 and 100 mg/kg doses (administered twice weekly). cTN3 PF had no adverse effects on the pre- and post-natal development of rats at doses up to 100 mg/kg (bi-weekly).

Summary: CDP870 binds to human TNFα with high affinity and cross-reacts with TNFα from non-human primates. However, CDP870 does not recognize TNFα from rodents. So it was not possible to conduct conventional reproductive toxicity studies with CDP870. Instead, the sponsor conducted Segment I (fertility and early embryonic development), Segment II (teratogenicity) and Segment III (pre- and post- natal development) using a homologous rat anti-TNF antibody (cTN3 γ1).

In the Segment I fertility and early embryonic development study in rats, male and female animals were treated with 20 and 100 mg/kg i.v. doses of cTN3, administered twice weekly. cTN3 had no effects on the fertility and early embryonic development in rats. In the segment II teratogenicity study with cTN3 in rats, the test agent was administered intravenously at 20 and 100 mg/kg doses on gestation days 1 and 4, or gestation days 6, 9, 13 and 16. cTN3 PF was not teratogenic in rats at the doses examined. In the segment III pre- and post- natal development study with cTN3 PF in rats, groups of pregnant females were intravenously administered the antibody at 0, 30 and 100 mg/kg doses (administered twice weekly). cTN3 PF had no adverse effects on the pre- and post-natal development of rats at doses up to 100 mg/kg (bi-weekly).

2.6.6.7 Local tolerance

Study title: CDP870 - Single-Dose Subcutaneous Local Tolerance Study in the Rat Followed by a 1, 7, 14 or 28-Day Observation Period.

Study Report No: 40000999

Site and testing facility:

Date Started: January 22, 1999

Date of Study Report: May 17, 2000

GLP compliance: Yes

QA- Report Yes (X) No ( ): Yes
Lot and batch numbers: CDP870, Batch # 12842/35
Species/strain: WI (Dlx/BRL/Han)BR rats.

Methods: Local tolerance of CDP870 was determined in rats (12 animals/sex/group) following single subcutaneous administration of 160 and 800 mg/kg doses, followed by a 1, 7, 14 and 28 day observation period. The dosing volumes were 0.1 and 0.5 ml for the low and high dose groups, respectively. The control group received the vehicle (acetate buffer). The animals were observed daily for clinical signs and mortality. The body weights were recorded before treatment and weekly thereafter. Food consumptions by individual animal were determined weekly. The injection site was assessed visually and by palpation daily. Three animals/sex from each group were killed after 1, 7, 14 and 28 day observation period. Injection sites and untreated control sites (except from the control group) from all animals were examined histologically. The following grading scheme was used:
Erythema of skin overlaying the injection site- Grade 0, no erythema; Grade 1, erythema apparent within 10 mm of site of penetration; Grade 2, erythema apparent >10 mm from site of penetration; Grade 3, erythema affecting entire outer aspect of flank.
Swelling- Grade 0, no swelling; Grade 1, slight swelling at the injection site; Grade 2, swelling at the injection site, skin displaced; Grade 3, generalized swelling of local musculature.
Hemorrhage- Grade 0, no hemorrhage; Grade 1, signs of hemorrhage adjacent to injection site; Grade 2, signs of marked hemorrhage at injection site; Grade 3, generalized hemorrhage of local musculature.
Necrosis- Grade 0, no sign of necrotic change; Grade 1, focus of necrosis apparent at injection site; Grade 2, necrosis apparent >5 mm from injection site; Grade 3, generalized necrotic changes of local musculature.

Toxicokinetics: At necropsy, samples of blood (1 ml) were collected from the abdominal aorta for determination of plasma CDP870 concentrations.

Results: No treatment-related clinical signs were observed in any group, and CDP870 had no adverse effects on body weight and food consumption of the animals. There was no observable reaction at the injection site at any time following administration of CDP870. When graded for erythema, swelling of the musculature, hemorrhage and necrosis, all animals scored zero on all occasions.

Histopathological examinations of animals given CDP870 and killed on day 2 showed evidence of minimal or slight edema of the subcutis, especially the pennniculus muscle. In some animals, there was minimal local hemorrhage associated with the edema. By day 8, there was reduced edema with a scattering of inflammatory cells, termed cellulitis. These reactions were slightly stronger in the high dose animals. The cellulitis resolved to normal skin by day 14 in all low dose animals and by day 29 in all high dose animals. Histological findings at the injection sites of animals on day 2 and day 8 are shown in the Table below.

Group incidence: histological findings - Day 2

Test article	Control	CDP870	
Group	1	2	3
Level (mg/kg)	0	160	800

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STUDY NUMBER: 106478

TABLE INCLUDES:
SEX=ALL;GROUP=ALL;WEEKS=ALL
DEATH=1;FIND=ALL;SUBSET=ALL

		--- NUMBER-OF-ANIMALS-AFFECTED ---					
	SEX:	-----MALE----			----FEMALE---		
	GROUP:	-1-	-2-	-3-	-1-	-2-	-3-
ORGAN AND FINDING DESCRIPTION	NUMBER:	3	3	3	3	3	3
** TOP OF LIST **							
LEFT FLANK	NUMBER EXAMINED:	3	3	3	3	3	3
--OEDEMA		0	3	3	0	2	3
--CELLULITIS		0	0	0	0	1	0
--HAEMORRHAGE		0	2	2	0	1	1
--MYOSITIS/MYOPATHY		1	1	0	0	0	0
--NEEDLE-TRACK LESION		0	0	0	0	0	2
RIGHT FLANK	NUMBER EXAMINED:	0	3	3	0	3	3
--MYOSITIS/MYOPATHY		0	0	0	0	1	0
** END OF LIST **							

Group incidence: histological findings - Day 8

Test article	Control	CDP870	
Group	1	2	3
Level (mg/kg)	0	160	800

PRINTED: 17-MAY-00
PAGE: 1

STUDY NUMBER: 108478

		--- NUMBER-OF-ANIMALS-AFFECTED ---					
TABLE INCLUDES:							
SEX=ALL;GROUP=ALL;WEEKS=ALL	SEX:	-----MALE----			----FEMALE---		
DEATH=2;FIND=ALL;SUBSET=ALL							
	GROUP:	-1-	-2-	-3-	-1-	-2-	-3-
ORGAN AND FINDING DESCRIPTION	NUMBER:	3	3	3	3	3	3
** TOP OF LIST **							
LEFT FLANK	NUMBER EXAMINED:	3	3	3	3	3	3
--CELLULITIS		0	1	3	0	1	1
--MYOSITIS/MYOPATHY		0	0	0	0	1	1
--DERMATITIS		0	0	0	0	0	1
RIGHT FLANK	NUMBER EXAMINED:	0	3	3	0	3	3
--MYOSITIS/MYOPATHY		0	1	0	0	0	0
** END OF LIST **							

Toxicokinetics: CDP870 was detected in the plasma of all animals administered the drug that were sacrificed on days 1, 7 and 14 post-dose. The plasma concentrations of the drug increased with increasing dose. The mean plasma concentrations were 393.1, 80.7, 6.3 and 0.4 μg/ml for animals receiving the 160 mg/kg dose, and 1167.3, 506.2, 42.2 and 0.5 μg/ml for animals receiving the 800 mg/kg dose, on days 1, 7, 14 and 28, respectively.

Summary: A single subcutaneous administration of CDP870 to rats at dose levels of 160 and 800 mg/kg was well-tolerated in rats without showing any clinical reaction at the injection site. On microscopic examination of the injection sites, minimal to slight edema with cellulitis (dose-dependent) was observed in CDP870-treated rats. These effects resolved by day 29 in all groups.

Study title: **PHA-738144 (CDP870): Single Dose Subcutaneous Local Tolerance Study in the Rat Followed by a 2, 8, 15 and 29 Day Observation Period.**

Study Report No: 40001362 (Study # SA5219)
Site and testing facility: GD Searle LLC, Skokie, IL.

Date Started: August 19, 2002

Date of Study Report: August 08, 2003

GLP compliance: Yes

QA- Report Yes (X) No ( ): Yes
Lot and batch numbers: PHA-738144, Lot # Cell014319, #19964 and #SP19991
Species/strain: ~~/I (Han) IGS BR rats.

Methods: The purpose of the study was to assess the local tolerance potential of two different active formulations of PHA-738144 (CDP870) following acute subcutaneous administration in rats. Six groups of animals (12 animals/sex/group) were administered the vehicle, placebo, current active formulation, lyophilized active reconstituted formulation, placebo or lyophilized active formulation by a single subcutaneous injection. Injections (100 mg/kg; 0.5 ml volume) were made on the left flank (L), and the right flank (R) was used for comparison only. The design of the experiment is summarized in the Table below.

5.1. Experimental Design

Test Group	Formulations (0.5 mL/dose)	PHA-738144 (mg/mL)	Number of Animals/Group Toxicology Males	Females
1	Vehicle	0	12	12
2	Lyophylized Placebo	0	12	12
3	Current Active Formulation	200	12	12
4[a]	Lyophylized Active (Reconstituted) Formulation	200	12	12
5	Lyophylized Placebo	0	12	12
6	Lyophylized Active (Reconstituted) Formulation	200	12	12

[a] Results received for the Lyophylized Active (Reconstituted) Analytical Entrance Assay on August 22, 2002 indicated the acceptable limits of ± ~~ were exceeded (26% or 148.2 mg/mL of the expected 200 mg/mL concentration). A Lyophylized Active (Reconstituted) Group 6 was added to achieve the target dose of 200 mg/mL and a Lyophylized Placebo Group 5 was added for comparison to the Lyophylized Active (Reconstituted).

The animals were observed twice daily for clinical signs and mortality. Body weights were measured prior to treatment and on days 1, 5 and weekly thereafter. Food consumption was measured pretreatment and once weekly thereafter. Injection sites were evaluated for erythema, swelling of the musculature, hemorrhage and necrosis, and graded from 0 to 3. Three animals/sex from each group were sacrificed on days 2, 8, 15 and 29, and gross examinations of the injection sites were conducted. A 3 X 3 cm area of the injection sites were also examined histologically.

Results: There were no mortalities or treatment-related clinical signs in any group. Body weights and food consumptions were comparable in all groups. Slight swelling and erythema at the injection sites were observed in all groups. None of the surviving animals had erythema or swelling on Day 8 post-injection. Swelling at the injection site was seen in 3/12, 2/12, 9/12, 5/12, 4/12 and 7/12 males in groups 1, 2, 3, 4, 5 and 6, respectively, indicating no apparent differences between the current formulation and the lyophilized active formulation. In females, swelling occurred in 0/12, 3/12, 2/12, 1/12, 5/12 and 10/12 animals from groups 1, 2, 3, 4, 5 and 6, respectively, indicating an increase in the local swelling with the lyophilized active formulation and the lyophilized vehicle as compared with the current formulation. Macroscopic examination of the injection sites showed mild edema at the subcutaneous injection sites of animals from groups 3, 4, 5 and 6. Histopathological changes at the injection site were seen in all groups including the control, and were characterized by lymphohistiocytic infiltrates, edema, myodegeneration and/or accumulation of vacuolated macrophages. There were no remarkable differences in the severity of histologic changes between the two formulations of CDP870.

Summary: Following single subcutaneous injections of the lyophilized product and the current formulation of CDP870 to rats, no difference in the clinical or morphologic end points of local tolerance were observed between the two formulations.

2.6.6.8 Special toxicology studies

No special toxicology studies were submitted under the BLA.

Other Studies:

CDP870: Single Dose Intravenous Range Finding Intravenous Toxicity Study in the Rat (Study Report # 40000905)

Methods: The study was conducted to determine the toxicity of CDP870, following a single intravenous administration of 0, 100, 200, 400 and 1000 mg/kg doses in rats (Han Wistar; 5 animals/sex/group). Groups 1, 2 and 3 were dosed on day1, group 4 on day 2 and group 5 on day 3. The dosing volume was 52.4 ml/kg. The animals were observed daily for clinical signs and mortality for 7 days after dosing. Body weights were measured before treatment on the first day of dosing, day 2 and before necropsy on day 8. Food consumption was determined daily. The animals were killed and discarded on day 8 without further examinations.

Results: No treatment-related clinical signs were observed in any group. The body weights and food consumptions of animals from all groups were similar during the observation period. Thus, single intravenous administration of CDP870 to rats, at doses up to 1000 mg/kg, was well-tolerated and without any effects.

CDP870: 5-Day Intravenous Range-Finding Toxicity Study in the Rat (Study Report # 1084/66-D6154).

Methods: The study was conducted to determine the toxicity of CDP870 following intravenous administration to the rat for 5 days. Groups of rats (5 animals/sex/group) were administered CDP870 at doses of 0, 50, 100 and 400 mg/kg (20.9 ml/kg) for 5 consecutive days. The animals were observed daily for clinical signs and mortality. Body weights were measured before treatment on the first day of dosing, day 5 and before necropsy. Food consumption was recorded daily. Blood samples for clinical pathology were obtained from all animals at necropsy on day 6. All animals were subjected to complete necropsy examinations. The weights of the following organs were recorded: adrenals, brain, heart, kidney, liver, ovaries, pituitary, prostate, spleen, testes+epididymides, and thyroids+parathyroids.
Histopathological examinations of the following organs were conducted: adrenals, aorta, brain, cecum, colon, duodenum, eyes, femur with bone marrow and articular surface, gross lesions, Harderian glands, heart, ileum, injection sites, jejunum, kidney, lacrimal glands, liver, lungs, mammary gland, lymph nodes, muscle, esophagus, optic nerves, ovaries, pancreas, pituitary, prostate, salivery glands, sciatic nerves, seminal vesicles, skin, spinal cord, spleen, sternum with bone marrow, stomach, testes+epididymides, thymus, thyroids+parathyroids, tongue, trachea, urinary bladder, uterus, Zymbal glands, vagina.

Blood samples for toxicokinetic analysis were taken from all animals on day 6, approximately 24 hours after the final dose.

Results: No treatment-related clinical signs or mortalities were observed in any group. Body weights and food consumptions were not affected by the treatment. Aspartate aminotransferase (AST) levels in the mid (22%) and high dose (18%) males and high dose females (27%) were slightly lower than that of controls. Alanine aminotransferase (ALT; 45% in males, 20% in females) and alkaline phosphatase (ALP; 50% in males, 20% in females) levels of the high dose animals were also lower than that of controls. A slight increase in the spleen weight was observed in male (14 to 21%) and female (26 to 36%) animals receiving the drug.

In a separate study (Study report #40001000), the sponsor conducted hematological examinations of rats treated with 50, 100 and 400 mg/kg intravenous doses of CDP870 for 5 days. Prothrombin time (PT) and activated partial thromboplastin time (APTT) were increased in the high dose males and females (13.2% and 12.8% for PT, and 39.5% and 47.1% for APTT, in males and females, respectively). High dose males and females had slight reductions of platelet (15.5% and 9.2% in males and females, respectively) and WBC (22.2% and 16.1% in males and females, respectively) levels.
High dose animals of both sexes showed histiocytic vacuolation in the hemolymphoreticular tissue, predominantly in the spleen and lymph nodes, and to a lesser extent in the bone marrow, lung, intestine and adrenal. In mid dose animals, there was a minor histiocytic vacuolation in the spleen of males and females and in the mandibular lymph node of one male. The change was characterized by variably sized vacuoles within the histiocytic cytoplasm. The incidences of histiocytic vacuolation for different groups are summarized in the Table below.

Incidences	Males				Females			
	Control	50 mg/kg	100 mg/kg	400 mg/kg	Control	50 mg/kg	100 mg/kg	400 mg/kg
Femur + Marrow -Histiocytic vacuolation	0/5	0/5	0/5	3/5	0/5	0/5	0/5	3/5
Sternum + Marrow -Histiocytic vacuolation	0/5	0/5	0/5	3/5	0/5	0/5	0/5	3/5
Spleen -Histiocytic vacuolation	0/5	0/5	4/5	5/5	0/5	0/5	4/5	5/5
Mesenteric lymph nodes -Hisriocytic vacuolation	0/5	0/5	0/5	5/5	0/5	0/5	0/5	5/5
Duodenum -Histiocytic vacuolation	0/5	0/5	0/5	5/5	0/5	0/5	0/5	3/5
Colon -Histiocytic vacuolation	0/5	0/5	0/5	3/5	0/5	0/5	0/5	3/5
Adrenal -Histiocytic vacuolation	0/5	0/5	0/5	3/5	0/5	0/5	0/5	2/5
Mandibular lymph node -Histiocytic vacuolation	0/5	0/5	1/5	5/5	0/5	0/5	0/5	5/5
Lung -Histiocytic vacuolation	0/5	0/5	0/5	4/5	0/5	0/5	0/5	1/5

CDP870 was detected in the plasma of all animals receiving the drug, and the plasma concentrations increased with increasing dose. The mean plasma CDP870 concentrations were 1.10, 1.89 and 5.95 mg/ml for 50, 100 and 400 mg/kg groups, respectively. No differences in the plasma levels were observed between males and females. Plasma concentrations of CDP870 in male and female animals from different groups are shown in the Table below.

CDP870 CONCENTRATIONS (µg/mL)
(Geometric Mean and 95% Confidence Interval)

GENDER		CDP870 DOSE (mg/kg)			
		0	50	100	400
MALE	MEAN	0.10	1109.57	1903.27	5901.73
	95% CI	0.100, 0.100	965.971,1274.518	1775.500,2040.228	4658.351,7476.977
	N	5	5	5	5
FEMALE	MEAN	0.10	1084.90	1876.84	5991.90
	95% CI	0.100, 0.100	935.253,1258.498	1724.491,2042.657	5529.985,6492.403
	N	5	5	5	5
ALL	MEAN	0.10	1097.17	1890.01	5946.64
	95% CI	0.100, 0.100	1014.377,1186.715	1811.555,1971.862	5401.661,6546.611
	N	10	10	10	10

Note: Values below the limit of quantification were set to 0.1 µg/mL.

Summary: In the 5-day intravenous dose-ranging study with CDP870 in rats, groups of animals (5 animals/sex/group) were administered the drug at doses of 0, 50, 100 and 400 mg/kg (20.9 ml/kg) for 5 consecutive days. A slight increase in the spleen weight was observed in male and female animals receiving the drug. High dose animals of both sexes showed histiocytic vacuolation in the hemolymphoreticular tissue, predominantly in the spleen and lymph nodes, and to a lesser extent in the bone marrow, lung, intestine and adrenal. A minor histiocytic vacuolation in the spleen was also observed in males and females receiving the mid dose. Thus, the 50 mg/kg/day dose was the no effect dose in this 5-day dose-ranging study.

2.6.6.9 Discussion and Conclusions

Toxicology studies with CDP870 were conducted in monkeys following administration of single and multiple doses. Intravenous administration of single doses up to 870 mg/kg was well-tolerated without any treatment-related adverse effects. In a 28-day i.v. toxicity study in monkeys, decreased hemoglobin, RBC and packed cell volume was observed immediately after administration of 50, 100 and 400 mg/kg weekly doses. Increased WBC levels were observed in male and female monkeys receiving 10 and 100 mg/kg subcutaneous doses for 13 and 26 weeks. Hematological parameters returned to their normal levels following a 13-week treatment-free recovery period. Histiocytic vacuolation in the hemolymphoreticular tissues (splenic red pulp, medullary sinuses of the mandibular and mesenteric lymph nodes, bone marrow, thymus) were observed in animals treated with the 400 mg/kg dose for 28 days. Vacuolation (foamy macrophages) was also observed in different organs (choroid plexus, adrenals, mesenteric and mandibular lymph nodes, lamina propria of the urinary bladder and endometrial stromal mucosa of the uterus and spleen) in monkeys receiving a 100 mg/kg weekly dose for 13, 26 or 52 weeks. Vacuolation of the hemolymphoreticular tissues may be related to the pharmacological effects of the drug. An increase in the activated partial thromboplastin time (APTT) was observed in monkeys receiving 50 and 100 mg/kg doses. A similar increase in the APTT was also observed in an *ex vivo* study with monkey blood. CDP870 was not genotoxic in a battery of genotoxicity assays. Segment I (fertility and early embryonic development), Segment II (teratogenicity) and Segment III (pre- and post- natal development) reproductive toxicology studies were conducted in rats using a [redacted] rat anti-TNF antibody (cTN3 [redacted] cTN3 had no effects on the fertility and early embryonic development, and it was not teratogenic in rats. cTN3 PF had no adverse effects on the pre- and post-natal development in rats at i.v. doses up to 100 mg/kg (bi-weekly). Thus, the toxicology studies conducted with CDP870 do not reveal any nonclinical safety issues relevant to its clinical use.

2.6.6.10 Tables and Figures

Tables and Figures are incorporated in appropriate sections of the review.

2.6.7 TOXICOLOGY TABULATED SUMMARY

N/A

2 Page(s) Withheld

_____ Trade Secret / Confidential

✓ Draft Labeling

_____ Deliberative Process

OVERALL CONCLUSIONS AND RECOMMENDATIONS

Tumor necrosis factor alpha (TNFα) is a multifunctional cytokine involved in the normal physiological response to infection. There is considerable evidence that excessive TNFα activity is involved in the pathogenesis of inflammatory bowel disease (IBD). In humans, TNFα is strongly expressed in bowel wall in areas affected by Crohn's disease, and fecal concentrations of TNFα have been shown to reflect clinical severity of the disease. Infiltrating leucocytes are the major source of TNFα. CDP870 (CIMZIA) is a recombinant, humanized antibody Fab' fragment with specificity for human TNFα. The sponsor submitted the licensing application for marketing the drug for inducing clinical response, maintaining clinical response and remission in patients with active Crohn's disease. The proposed dose of CIMZIA for Crohn's disease patients is 400 mg given as two s.c. injections at weeks 0, 2 and 4, followed by a maintenance regimen of 400 mg every 4 weeks.

In support of the current BLA, the sponsor submitted the following preclinical studies with CDP870: Pharmacology; Absorption, Distribution, and Excretion studies in monkeys and rats; Toxicology- single dose intravenous tolerance/toxicity study in the monkey, repeated dose toxicity studies: 28-day intravenous toxicity study in the monkey, 13- and 26-week subcutaneous toxicity study in the monkey, 52-week subcutaneous toxicity study in the monkey; Genotoxicity- bacterial reverse mutation (Ames assay) assay, chromosomal aberration assay in human peripheral blood lymphocytes, mouse bone marrow micronucleus assay; Reproductive and Developmental Toxicity- Intravenous Segment I fertility and reproductive performance study in rats with anti-rat TNFα monoclonal antibody equivalent to CDP870, Intravenous Segment II teratogenicity study in rats with anti-rat TNFα monoclonal antibody equivalent to CDP870, Intravenous Segment III pre- and post- natal development study in rats with anti-rat TNFα monoclonal antibody equivalent to CDP870; Local Tolerance- single dose subcutaneous local tolerance studies in the rat; Other Studies- single-dose range finding i.v. toxicity study in the rat; 5-day i.v. range finding toxicity study in the rat.

CDP870 was found to bind to recombinant human TNFα with high affinity (K_d, about 90 pM). It was also found bind to cynomolgus monkey TNFα with a low affinity (about $1/40^{th}$ of that of human TNFα), but did not bind to rat, mouse, guinea pig and rabbit TNFα. The binding affinity of CDP870 for human TNFα (affinity, 90.2±14.3 pM) was higher than that of infliximab (affinity, 228.7±39.1 pM) and adalimumab (affinity, 158.3±23.1 pM) and lower than that of etanercept (affinity, 33.2±6.2 pM). It neutralized soluble and membrane TNFα, inhibited binding of TNFα to human TNF receptors p55 and p75 (types I and II) and inhibited LPS-induced cytokine production in human monocytes. In addition, CDP870 neutralized the biological activity of human TNFα *in vivo*, in animals in which human TNFα was the physiologically active molecule. Intravenously administered CDP870 inhibited human TNFα-induced neutrophil accumulation in the peritoneal cavity of mice and pyrexia in rabbits, and inhibited chronic inflammatory polyarthritis in transgenic mice. It did not mediate antibody-dependent cell-mediated cytotoxicity, and it is therefore not cytotoxic to TNF-expressing cells.

PEGylation of HTNF 40 IgG with the 40 KD PEG increased the elimination half-life ($t_{1/2\alpha}$) of the compound in monkeys following a single i.v. dosing. The plasma exposure level for the 40

KD PEGylated IgG (CDP870) was higher than that for the ~ 3D PEGylated IgG (78% and 30% of the non-PEGylated IgG, respectively). PEGylation also decreased the immunogenicity of the antibody. CDP-870 also showed a longer half-life and higher plasma exposure levels than that of Fab' in rats following an i.v. dose. Following subcutaneous administration of single doses (3 and 31 mg/kg) of CDP870 to monkeys, plasma concentrations increased with increasing dose, and the maximum plasma concentrations were reached between 24 and 48 hours with a $t_{1/2}$ of about 200 hours (8.4 days). Following subcutaneous administration (10 and 200 mg/kg, twice a week for 4 weeks) in rats, the C_{max} values were reached in 24 hours, and the $t_{1/2}$ value was about 60 hours. The estimated bioavailability in rats following s.c. administration was 23.5% in males and 33.8% in females. Tissue distribution of CDP870 in rats following an i.v. dose was similar to that of the non-PEGylated form. At 3 hours following administration, the highest level was found in the kidneys, followed by lung, liver and spleen. In humans, the peak CDP870 concentrations were attained between 54 and 171 hours following s.c. administration. The terminal elimination half-life ($t_{1/2}$) was approximately 14 days for all doses tested. Following s.c. administration to humans for 12 weeks, anti-CDP870 antibodies were detected in 5% (at 800 mg/4 week dose) to 67% (at 50 mg/4 week dose) of the subjects, depending on the doses administered. The presence of the antibody decreased the C_{max} and AUC by more than 50%. CDP870 was not an inhibitor of P-glycoprotein.

Single intravenous administration of CDP870 at doses up to 870 mg/kg, was well tolerated in cynomolgus monkeys,. Treatment-related effects on B and T lymphocytes were observed at all doses. However, no control group was included in the study.

In the 28–day intravenous toxicity study with CDP870 in cynomolgus monkeys, the drug was administered at doses of 0, 50, 100 and 400 mg/kg at weekly intervals. Treatment group animals had decreased hemoglobin, RBC and packed cell volume immediately following infusion and continued throughout the treatment period. Vacuolated histiocytes were observed at the injection sites of the high dose animals. The high and mid dose animals of both sexes had histiocytic vacuolation in the hemolymphoreticular tissue, notably in the splenic red pulp and in the medulary sinuses of the mandibular and mesenteric lymph nodes. Vacuolated histiocytes were also observed to a lesser extent in the bone marrow, thymus, carotid plexus and ovary. The 50 mg/kg/week dose was the no effect dose, and the target organs of toxicity were the hemolymphoreticular system, carotid plexus and the ovary.

In the 13/26–week subcutaneous toxicity study with CDP870 in cynomolgus monkeys, the drug was administered to groups of animals at doses of 0, 10 and 100 mg/kg, once weekly. At the end of the 13 or 26 weeks treatment period, 2 animals/sex/group were left untreated for a 13-week recovery period. There were no treatment-related changes in clinical signs, body weight, ECG, gross pathology, histopathology or clinical chemistry parameters in any group. Foamy macrophages in different organs were observed in animals receiving the 100 mg/kg weekly dose for 13 or 26 weeks. The changes were not completely reversible at the end of the 13-week recovery period. Foamy macrophages at the injection sites were observed in animals receiving both 10 and 100 mg/kg doses. The presence of foamy macrophages at the injection sites was reversible in the 10 mg/kg dose group but not in the 100 mg/kg dose group. Anti-CDP870 antibodies were detected in week 9 in two females receiving the 10 mg/kg

dose, and these animals were sacrificed. Six more animals had low levels of anti-CDP870 antibodies at the end of the dosing period. The 10 mg/kg weekly dose was the no effect dose.

In a 52-week subcutaneous toxicology study with CDP870 in cynomolgus monkeys, the drug was administered at 50 and 100 mg/kg doses, once weekly. Although, the sponsor stated that half of the animals were left untreated for a 26-week recovery period, no data were provided for the recovery groups in this interim study report. An increase in the activated partial thromboplastin time (APTT) was observed in males and females receiving CDP870. Animals treated with CDP870 at 100 mg/kg/week showed vacuolation in several organs, including sinusoidal histiocytes, submandibular and mesenteric lymph nodes, splenic red pulp, zona reticularis of the adrenal gland, lamina propria of fallopian tube, uterine body and cervix, choroid plexus of the brain and the subcutaneous tissue of the injection site. Vacuolation was also observed in some organs at the 50 mg/kg/week dose. Following s.c. administration to monkeys, the plasma exposure levels of CDP870 increased with increasing dose with no differences between males and females. Antibodies to CDP870 were detected in two animals receiving the 50 mg/kg dose.

The genotoxic potential for CDP870 was assessed in the bacterial reverse mutation assay (Ames assay), the human peripheral blood lymphocyte chromosomal aberration assay and the mouse bone marrow micronucleus assay. CDP870 had no genotoxic potential in any of these assays.

CDP870 binds to human TNFα with high affinity and cross-reacts with TNFα from non-human primates. However, CDP870 does not recognize TNFα from rodents. So it was not possible to conduct conventional reproductive toxicity studies with CDP870. Instead, the sponsor conducted Segment I (fertility and early embryonic development), Segment II (teratogenicity) and Segment III (pre- and post- natal development) using a homologous rat anti-TNF antibody (cTN3 γ1).

In the Segment I fertility and early embryonic development study in rats, male and female animals were treated with 20 and 100 mg/kg i.v. doses of cTN3, administered twice weekly. cTN3 had no effects on the fertility and early embryonic development in rats. In the segment II teratogenicity study with cTN3 in rats, the test agent was administered intravenously at 20 and 100 mg/kg doses on gestation days 1 and 4, or gestation days 6, 9, 13 and 16. cTN3 PF was not teratogenic in rats at the doses examined. In the segment III pre- and post- natal development study with cTN3 PF in rats, groups of pregnant females were intravenously administered the antibody at 0, 30 and 100 mg/kg doses (administered twice weekly). cTN3 PF had no adverse effects on the pre- and post-natal development of rats at doses up to 100 mg/kg (bi-weekly).

A single subcutaneous administration of CDP870 to rats at dose levels of 160 and 800 mg/kg was well-tolerated in rats without showing any clinical reaction at the injection site. On microscopic examination of the injection sites, minimal to slight edema with cellulitis (dose-dependent) was observed in CDP870-treated rats. These effects resolved by day 29 in all groups.

Conclusions:

CDP870 is a humanized antibody Fab' fragment with specificity for human TNFα, which is manufactured in *E. coli* and then conjugated to ——— polyethylene glycol (PEG) ———. CDP870 binds to human TNFα with high affinity *in vitro*, and neutralizes the biological activity of TNFα *in vitro* and *iv vivo*. The sponsor submitted BLA 125160 for use of CDP870 for the treatment of Crohn's disease.

CDP870 binds to human TNFα with high affinity and weakly cross-reacts with TNFα from non-human primates. However, CDP870 does not recognize TNFα from rodents. So, toxicity studies with CDP870 were conducted in cynomolgus monkeys. In repeat dose toxicity studies in monkeys, slight hematological changes (decreased hemoglobin, RBC and packed cell volume, increased WBC) were observed and these changes were reversible. In addition, vacuolation of several tissues, particularly hemolymphoreticular tissues were observed in animals receiving high doses. This may be related to the pharmacological actions of the drug. About 5% of the animals receiving i.v. or subcutaneous doses of CDP870, developed anti-CDP870 antibodies. CDP870 was not genotoxic in a battery of genotoxicity assays. As CDP870 does not cross-react with TNFα from rodents, reproductive toxicity studies (Segment I fertility and early embryonic development, Segment II teratogenicity and Segment III pre- and post- natal development) were conducted in rats using a homologous rat anti-TNF antibody (cTN3 γ1). cTN3 had no effects on the fertility and early embryonic development, it was not teratogenic and had no effects on pre- and post- natal development in rats. Thus, the preclinical studies with CDP870 suggest that the sponsor's proposed dose of the product appears to be safe for the treatment of patients with Crohn's disease.

Unresolved toxicology issues (if any): None

Recommendations:

The sponsor conducted adequate preclinical studies with CDP870 to determine the safety of the drug, and the sponsor's proposed dose appears to be safe for the proposed indication. Thus, from a preclinical standpoint, the BLA application is approvable.

Suggested labeling: See the labeling part of the review.

Sushanta Chakder 10/31/06

Sushanta Chakder, Ph. D. Date
Pharmacologist, HFD-180

Comments: Concur

Jasti B. Choudary 10/31/06

Jasti B. Choudary, Ph.D., B. V. Sc. Date
Supervisory Pharmacologist, HFD-180

cc.
BLA
HFD-180
HFD-181/CSO
HFD-180/Dr. Chakder
HFD-180/Dr. Choudary
HFD-102/Dr Jacobs
HFD- 048/Dr. Viswanathan

R/D Init.: J. Choudary 10/23/06