Fecal Analysis: Fecal samples were collected on during on days 184 and 185 (week 27) from fasted animals in control, LD, MD, HD and pair-fed control groups scheduled for terminal sacrifice and on day 186 from non-fasted animals in the same groups scheduled for recovery sacrifice. The samples were analyzed for fat, protein (% nitrogen), and pH. Dry weight of each sample was also recorded.

Hormone Analysis: Blood samples were collected from each animal in control, LD, MD, HD and pair-fed control (except those designated for PK analysis) at scheduled sacrifices. Serum harvested from the blood was analyzed for TSH, T4, T3, LH and PTH.

Vitamin D Analysis: Recovery animals were fasted overnight during week 28 and bled before recovery sacrifice. Serum harvested was analyzed for 1, 25-dihydroxyvitamin D.

Lymphocyte Subsets: To determine the immunotoxic potential of this compound, animals designated for recovery were fasted overnight during week 27 and whole blood was collected for lymphocyte subset analysis. A portion of spleen and thymus were also collected from animals in control, LD, MD, HD and pair-fed control at terminal sacrifice for lymphocyte subset analysis.

Results:

Mortality: No test material-related deaths. Sponsor stated that 2/30 (0 mg/kg), 2/40 (MD) and 1/48 (HD) died as a result of gavage error. Histopathology data was not provided.

Clinical signs: Incidence

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
n	30		48		48		48		30	
Sex	M	F	M	F	M	F	M	F	M	F
Swollen neck			1		2		2			
Swollen paws					2		3			
Scaly tail			1		3	10	22	22		
Scaly tail tip		1	5		4	5	4	3		2
Wart-like lesions on tail		1	1	7	5	4	11	8		1

Empty cells indicate zero incidence

Body weights: (g)

Table showing decrement and % decrement in mean body weight gain

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
Day -4	125	122	124	119	124	119	124	118	124	115
Day 183	657	392	604*	416	580*	397	506*	356	592*	362
Decrement	0	0	53	-	77	-	151	36	65	30
% decrement	0	0	8	-	12	-	23	9	10	8
Day 218 (Recovery)	640	371	589	419	586	405	531*	341	599	348
Decrement	0	0	51	-	53	-	109	30	41	23
% decrement	0	0	8	-	10	-	17	8	6	6

*$p < 0.05$

Food consumption: (g)

Table showing decrement and % decrement in mean food consumption

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
Week -1	111	113	112	110	112	111	115	115	111	107
Week 26	184	138	176	154	170	151	160*	138	156*	129
Decrement	0	0	8	-	14	-	24	0	28	9
% decrement	0	0	4	-	8	-	13	0	15	7
Week 30 (Recovery)	178	141	188	161	180	158	174	137	169	128
Decrement	0	0	-	-	-	-	4	4	9	13
% decrement	0	0	-	-	-	-	2	3	5	9

*$p < 0.05$

Ophthalmoscopy:

WEEK 26: SUMMARY OF OPHTHALMIC OBSERVATIONS

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
Choroidal atrophy, OD			1/48		1/48	1/48	2/48			1/30
Dacryoadenitis, OS		3/30	1/48				2/48	1/48	1/30	1/30
Dacryoadenitis, OD			1/48							1/30
Corneal dystrophy, OD	1/30									
Corneal dystrophy,OS						1/48	1/48			
Corneal dystrophy,OU		1/30				1/48	2/48	1/48		
Anterior synechia, OS							1/48			
Choroidal hemorrhage, OS								1/48		
WEEK 30: RECOVERY OPHTHALMIC OBSERVATIONS										
Inflammation of iris, OD			2/48							
Inflammation of cornea, OS							1/48			
Retinal hemorrhage, OD							1/48			
Chronic dacryoadenitis, OS		2/30						1/48		
Chronic dacryoadenitis, OD		1/30								
Pthisis bulbi, OD		1/30						1/48		
Inflammation of iris, OD										1/30

Empty cells indicate zero incidence

Electrocardiography: No data.

Hematology:

WEEK 27 DATA

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
RBC (E6/UL)		7.8		7.5		7.3*		7.2*		7.7
HGB (g/dl)	15.4	15.5	14.7	14.7	14.6*	14.4*	14.7	14.0*	15.5	15.2
HCT (%)		42.8		41.1		40.3		39.2*		42.2
PLT (E3/UL)	1012	953	983	883	934	792*	787*	796*	981	883
WBC (E3/UL)		4.7		6.5		7.4*		8.0*		4.2
LYMPH (E3/UL)		4.0		5.6		5.7		6.5*		3.6
N-SEG (E3/UL)	1.5		2.2		6.0*		3.5*		1.4	

*** $p < 0.05$**

At the end of the recovery period (week 32) all hematology parameters had returned to normal limits.

Clinical chemistry:

WEEK 27 DATA

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
T. PRO (g/dl)	7.4		7.1		7.1		6.9*		7.1	
ALB (g/dl)	4.8		4.3*		4.1*		4.1*		4.6	
Bile acids (umol/l)	10		14		14		31*		14	
AST (IU/L)	139		170		199		301*		148	
ALT (IU/L)	38		45		69*		114*		36	
ALKP (IU/L)	77		49*		59*		50*		77	
CHOL (mg/dl)		95 ± 17		114		141 ± 59		134* ± 39		102
Electrophoresis ALB (g/dl)	5.0		4.6*		4.4*		4.5*		4.9	
α-1 globulin (g/dl)	0.2		0.5		0.7*		0.3		0.2	
γ-blobulin (g/dl)	0.4	0.6	0.2*	0.4 ± 0.10	0.2*	0.4 ± 0.21	0.3	0.3*	0.3	0.5
WEEK 31 (RECOVERY) DATA										
ALB (g/dl)	4.8		4.6		4.5*		4.3*		4.7	

GLU (g/dl)		121		118		116		104*		129

SD was recorded in a few cases for clarification of statistical significance; * p < 0.05

Urinalysis:

WEEK 27 DATA

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
Ca EXC (mg)	1.1	2.2	1.8	4.6*	2.2*	5.2*	2.9*	4.4*	1.1	2.1
Cl EXC (mmol)	0.5		0.7		0.8*		0.8*		0.5	
Urine Ca (mg/dl)		7.4		14.4*		14.0*		9.6		7.1
At the end of the recovery period (week 31), all urinalysis parameters had returned to normal limits.										

EXC = excretion; * p < 0.05

Organ weights:

WEEK 27 DATA

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
Epididymides (g)	1.56		1.24*		1.32*		1.15*		1.55	
Liver (g)	17.39		17.39		17.44		14.04*		14.23*	
Liver (%)	2.81	2.64	3.16*	3.15*	3.40*	3.33*	3.02	3.35*	2.63	2.80
Heart (g)	1.76		1.67		1.51*		1.52*		1.58	
Spleen (g)	0.82		0.66		0.71		0.54*		0.65	
Stomach (g)	2.31		2.23		2.12		1.92*		2.04	
Pituitary (g)	0.017	0.024	0.018	0.037*	0.016	0.034*	0.015*	0.033	0.018	0.025
Pituitary (%)		0.006		0.010*		0.009*		0.010*		0.008
Small intestine (%)	1.42	2.04	1.64*	2.28	1.69*	2.35	1.85*	2.60*	1.47	2.17
Large intestine (%)	0.80	1.05	0.86	1.07	0.90*	1.17	0.94*	1.28*	0.80	1.03
Kidney (%)	0.62		0.68		0.75*		0.70		0.61	
Cecum (%)	0.230	0.306	0.258	0.341	0.261*	0.352	0.313*	0.408*	0.263	0.327
Brain (%)	0.37		0.41		0.44*		0.48*		0.42*	
WEEK 32 (RECOVERY DATA)										
Epididymides (g)	1.50		1.39		1.41		1.30*		1.51	
Pituitary (g)		0.025		0.031		0.032		0.033*		0.024
Pituitary (%)	0.0026	0.007	0.003 1*	0.008	0.003*	0.008	0.003*	0.011*	0.0028	0.007 4
Small intestine (%)	1.25		1.39		1.51*		1.49*		1.27	
Large intestine (%)	0.73		0.750		0.80		0.87*		0.72	
Kidney (%)	0.60		0.68		0.65		0.71*		0.60	
Brain (%)	0.38		0.40		0.40		0.44*		0.40	

Absolute wt (g); Relative wt (%) = relative to body wt; * p < 0.05

Gross pathology:

WEEK 27 DATA

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
Kidneys Large pelvis(es)					1/10		1/10			
Kidneys Cysts								1/10		
WEEK 32 (RECOVERY DATA)										
Epididymides Small							1/10			
Testes Small			1/10				1/10			

Empty cells indicate zero incidence

Histopathology: Please note that sponsor did not provide severity scores for some lesions.

INTERIM SACRIFICE (WEEK 14)

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
Pancreas - apoptosis Individual cell necrosis							1/5(1)	1/5(1)		
Liver Coagulative necrosis							1/5			
Stomach – Chief cells Cytoplasmic vacuolation							1/5(1)	1/5(1)		
Mucosal necrosis					1/5(2)					
Mesenteric L. node Lymphoid depletion							1/5			
Lungs Alveolar histiocytosis								1/5		
Kidney Mineralization, tubular		1/5(1)						3/5(1)		
Renal tubular dilatation								1/5(1)		

1 = slight; 2 = mild; 3 = moderate; 4 = marked; 5 = severe

TERMINAL SACRIFICE (WEEK 27)

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
Esophagus Fibrosis, serosal							1/15	1/14		
Large intestine Mucosal necrosis							1/15			
Pancreas Acinar cell vacuolation							1/15(1)			
Salivary gland Cytokaryomegaly, seromucus acinar cells							1/15			
Stomach, chief cells Cytoplasmic vacuolation							2/15 1/15(1) 1/15(2)			
Epididymides Hypospermia							3/15			
Testis Degeneration/atrophy			1/15(1)		1/15(4)		3/15 1/15(2) 2/15(3)		1/15 (4)	
Mesenteric L. node Lymphoid depletion							1/15			
Skin, tail Hyperkeratosis, acantho -sis, pyogranulomatous dermatitis							8/15			
Eye Posterior synechia							1/15			
Caratact, sub-capsular							1/15			
Kidney Chronic progressive nephropathy	3/13 1/13(1) 2/13(2)	1/15(1)	13/15 7/15(1) 5/15(2) 1/15(3)	8/15 5/15(1) 3/15(2)	12/15 5/15(1) 5/15(2) 2/15(3)	9/15 5/15(1) 3/15(2) 1/15(3)	12/15 4/15(1) 4/15(2) 1/15(3)	4/14 3/15(1) 1/15(2)		1/15 (1)

1 = slight; 2 = mild; 3 = moderate; 4 = marked; 5 = severe

RECOVERY SACRIFICE (WEEK 32)

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
Testes Degeneration/atrophy			1/10(4)				2/10(4)			
Kidney Chronic progressive	2/9(1)						6/10 3/10(1)	1/10(1)		

nephropathy							3/10(2)			
Interstitial nephritis								1/10(1)		

1 = slight; 2 = mild; 3 = moderate; 4 = marked; 5 = severe

Sperm evaluation:

WEEK 27: SUMMARY OF SPERM EVALUATION

Dose (mg/kg/d)	0	300	600	1200	Pair fed control
Sperm motility					
Total motile sperm	170 ± 69	59 ± 55	79 ± 62	50 ± 59	157 ± 63
Percent motility	41 ± 11	23 ± 16	27 ± 15	18 ± 18	42 ± 10
Sperm concentration (10^6)	11 ± 3	6 ± 2	8 ± 5	5 ± 3	12 ± 3
Sperm Morphology					
Normal	98 ± 2	60 ± 21	75 ± 23	55 ± 28	98 ± 2
Amorphous	0 ± 0	3 ± 3	2 ± 4	10 ± 26	0.1 ± 0.4
Decapitated head	2 ± 2	36 ± 20	23 ± 22	33 ± 22	1 ± 2
Abnormal sperm (%)	2 ± 2	40 ± 21	26 ± 23	45 ± 28	2 ± 2
WEEK 32 (RECOVERY): SUMMARY OF SPERM EVALUATION					
Sperm motility					
Total motile sperm	162 ± 60	133 ± 38	138 ± 65	91 ± 74	201 ± 49
Percent motility	45 ± 7	45 ± 6	42 ± 11	31 ± 22	49 ± 5
Sperm concentration (10^6)	12 ± 4	8 ± 3	7 ± 2	5 ± 4	12 ± 2
Sperm Morphology					
Normal	98 ± 2	88 ± 9	86 ± 13	64 ± 36	98 ± 3
Amorphous	0 ± 0	1 ± 2	0.1 ± 0.3	2 ± 3	0.1 ± 0.3
Decapitated head	2 ± 2	10 ± 10	10 ± 11	26 ± 29	2 ± 3
Abnormal sperm (%)	2 ± 2	12 ± 9	14 ± 13	36 ± 36	3 ± 3

Hormone Analysis:

WEEK 27 DATA

Dose (mg/kg/d)		0		300		600		1200		Pair fed control	
Sex		**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**
Thyroxine (µg/dl)	M	4.2	2.2	3.1*	2.0	2.4*	2.0	3.2*	2.8*	3.3	2.2
	SD	0.56	0.40	1.007	0.51	0.83	0.56	1.07	0.55	0.79	0.54
Triiodothyronine (ng/ml)	M	73.4	84.4	62.4	85.7	61.6	80.2	51.5*	90.2	55.5*	80.6
	SD	11	15	19	14	12	18	8	18	12	26
WEEK 32 (RECOVERY DATA)											
At the end of the recovery period, T3 and T4 in treated males were not significantly different from controls. No T3 and T4 data was provided for recovery females.											

*** p < 0.05**

Bone Mineral analysis:

WEEK 27 DATA

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**
Dry Bone wt (g)	0.82		0.74*		0.71**		0.66*		0.75	
Dry bone weight of treated females were not statistically significantly different from control. None of the bone minerals analyzed (Ca, P) were statistically significantly different from control.										

*** p < 0.05**

Fecal Analysis:

WEEK 27 DATA

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**
Nitrogen (%)	3.70	3.74	3.81	3.74	4.05*	3.78	4.17*	3.89	4.32*	4.32*
Fat (%)	4.6		4.9		4.9		5.0*		5.0*	
Fecal wet weight, dry weight and pH were not statistically significantly different from control. * p < 0.05										

Serum 1, 25-Dihydroxyvitamin D (pg/ml):

WEEK 28 DATA

Dose (mg/kg/d)	0		300		600		1200		Pair fed control	
Sex	M	F	M	F	M	F	M	F	M	F
1, 25-Dihydroxyvitamin D		11.5		7.66		6.33*		7.44		11.6
1, 25-Dihydroxyvitamin D was statistically significantly decreased only in MD females. Differences in males did not achieve statistical significance.										

* $p < 0.05$

Lymphocyte subsets:

Lymphocyte Subsets (Absolute Numbers) in Rat Peripheral Whole Blood
Absolute Cell Number ($X10^3$)/mm^3 of Blood (MEAN ± SD)

FEMALE

LYMPHOCYTE SUBSETS	CONTROL (n=8)	PAIR-FED CONTROL (n=9)	1200 mg/kg/d (n=8)
Total Lymphocytes	4.1 ± 1.9	4.4 ± 1.2	6.0 ± 2.0
CD45RA+	1.4 ± 0.9	1.3 ± 0.4	2.1 ± 0.9
Pan T+	2.4 ± 1.0	2.7 ± 0.8	3.4 ± 1.1
CD3+	2.3 ± 0.9	2.7 ± 0.8	3.4 ± 1.1
CD3+CD4+	1.5 ± 0.6	1.8 ± 0.6	2.6 ± 0.8*
CD3+CD8+	0.8 ± 0.4	0.9 ± 0.4	0.8 ± 0.3

MALE

LYMPHOCYTE SUBSETS	CONTROL (n=9)	PAIR-FED CONTROL (n=8)	1200 mg/kg/d (n=8)
Total Lymphocytes	5.9 ± 1.9	6.4 ± 1.9	8.3 ± 2.8
CD45RA+	2.1 ± 0.7	2.4 ± 0.7	3.6 ± 1.1*
Pan T+	3.1 ± 1.2	3.3 ± 1.1	3.7 ± 1.6
CD3+	3.0 ± 1.2	3.3 ± 1.1	3.6 ± 1.6
CD3+CD4+	1.8 ± 0.7	2.0 ± 0.4	2.6 ± 1.0
CD3+CD8+	1.3 ± 0.6	1.3 ± 0.7	1.4 ± 0.6

* Absolute cell numbers were calculated by multiplying the "corrected" lymphocyte subset percentages by the absolute lymphocyte cell count.

Abbreviations and Symbols:
n = The number of animals included in analysis
* = p≤ 0.05

Lymphocyte Subsets (Absolute Numbers) In Rat Spleen
Absolute Cell Number ($X10^7$)/Spleen (MEAN ± SD)[a]

FEMALE

LYMPHOCYTE SUBSETS	CONTROL (n=5)	PAIR-FED CONTROL (n=5)	300 mg/kg/d (n=5)	600 mg/kg/d (n=5)	1200 mg/kg/d (n=5)
Total Leukocytes	23.9 ± 5.2	14.6 ± 3.3*	21.7 ± 7.1	26.2 ± 14.3	19.6 ± 10.2
CD43+	9.3 ± 2.3	5.5 ± 2.0*	9.7 ± 3.2	8.8 ± 4.5	7.8 ± 5.3
CD3+	10.7 ± 1.7	6.5 ± 1.6*	10.8 ± 3.4	10.7 ± 5.5	9.8 ± 6.2
CD3+CD4+	5.8 ± 1.5	3.4 ± 1.0*	6.7 ± 2.5	6.4 ± 3.2	6.1 ± 4.1
CD3+CD8+	4.2 ± 0.8	2.2 ± 0.6*	3.4 ± 0.8	3.1 ± 1.7	2.2 ± 1.4
IgM+	11.2 ± 2.4	6.6 ± 1.4*	9.7 ± 4.0	11.9 ± 7.2	8.9 ± 4.7

MALE

LYMPHOCYTE SUBSETS	CONTROL (n=4)	PAIR-FED CONTROL (n=5)	300 mg/kg/d (n=5)	600 mg/kg/d (n=5)	1200 mg/kg/d (n=5)
Total Leukocytes	23.4 ± 7.9	21.4 ± 9.6	26.7 ± 13.6	21.7 ± 1.8	22.0 ± 8.0
CD43+	8.4 ± 2.5	6.9 ± 1.6	11.2 ± 5.8	7.7 ± 1.3	8.1 ± 3.3
CD3+	9.7 ± 3.5	9.4 ± 3.4	12.7 ± 6.2	9.6 ± 1.6	10.2 ± 4.4
CD3+CD4+	5.8 ± 2.1	5.2 ± 0.8	7.7 ± 5.4	6.5 ± 2.0	6.8 ± 3.
CD3+CD8+	3.3 ± 1.8	2.9 ± 1.2	3.5 ± 1.9	2.1 ± 0.2	2.4 ± 0.7
IgM+	11.8 ± 4.1	11.9 ± 8.3	11.0 ± 5.6	10.3 ± 1.1	11.4 ± 4.3

[a] Absolute cell numbers were calculated by multiplying the "corrected" lymphocyte subset percentages by the absolute lymphocyte cell count.

Abbreviations and Symbols:
n = The number of animals included in analysis
* = p ≤ 0.05

Lymphocyte Subsets (Absolute Numbers) in Rat Thymus
Absolute Cell Number ($X10^7$)/Thymus (MEAN ± SD)

FEMALE

LYMPHOCYTE SUBSETS	CONTROL (n=3)	PAIR-FED CONTROL (n=4)	300 mg/kg/d (n=3)	600 mg/kg/d (n=3)	1200 mg/kg/d (n=5)
Total Leukocytes	14.1 ± 11.0	16.3 ± 13.3	12.2 ± 1.7	30.1 ± 6.9	15.6 ± 10.2
CD43+	13.3 ± 10.5	17.6 ± 14.1	11.6 ± 1.6	28.5 ± 6.5	14.7 ± 9.8
CD3+	11.4 ± 9.1	12.9 ± 10.2	9.7 ± 1.0	20.5 ± 6.5	12.5 ± 8.3
CD3+CD4+	11.2 ± 8.9	12.5 ± 9.9	9.5 ± 1.0	20.1 ± 6.4	12.2 ± 8.1
CD3+CD8+	10.6 ± 8.1	12.0 ± 9.3	9.2 ± 0.9	19.9 ± 5.9	11.5 ± 7.7
IgM+	0.7 ± 0.3	0.9 ± 0.6	0.5 ± 0.2	1.2 ± 0.4	0.6 ± 0.4
CD4+CD8-	0.6 ± 0.4	0.8 ± 0.5	0.6 ± 0.2	1.4 ± 0.5	1.0 ± 0.6
CD4+CD8+	0.7 ± 0.5	0.8 ± 0.6	0.5 ± 0.2	1.3 ± 0.3	0.9 ± 0.6
CD4+CD8+	12.6 ± 9.9	14.2 ± 11.9	10.9 ± 1.3	25.7 ± 5.9	13.4 ± 9.2
CD4+CD8-	0.1 ± 0.1	0.2 ± 0.1	0.2 ± 0.1	0.3 ± 0.2	0.1 ± 0.1

[a] Absolute cell numbers were calculated by multiplying the "corrected" lymphocyte subset percentages by the absolute lymphocyte cell count.

Abbreviations and Symbols:
n = The number of animals included in analysis

Toxicokinetics:

Dose (mg/kg/d)	300 = 100 mg/kg TID		600 = 200 mg/kg TID		1200 = 400 mg/kg TID	
Sex	M	F	M	F	M	F
Day 1 C_{max} (µg/ml)	7.78	6.91	11.1	9.07	14.4	12.8
$AUC_{0-8\ hr}$ (µg.h/ml)	15.6	14.6	33.7	30.0	41.4	47.0
T_{max} (h)	1.0	1.0	1.0	1.0	1.0	1.0
Week 13 C_{max} (µg/ml)	9.65	9.42	11.1	22.3	17.4	34.0
$AUC_{0-8\ hr}$ (µg.h/ml)	22.3	22.9	39.8	46.3	59.9	72.1
T_{max} (h)	1.0	1.0	1.0	1.0	1.0	1.0
Week 26 C_{max} (µg/ml)	12.4	10.2	18.2	25.2	22.4	26.7
$AUC_{0-8\ hr}$ (µg.h/ml)	30.7	26.1	55.9	40.2	68.0	62.7
T_{max} (h)	1.0	0.5	1.0	1.0	1.0	1.0

Summary of Study Findings:

In this rat study, animals were dosed orally by gavage with OGT 924 at 300, 600 and 1200 mg/kg/d (administered as three equally divided doses at 8 hr intervals) for 26 week followed by a 4-week recovery period. Dose-related increase in scaly tail was observed in both sexes. Wart-like lesions on the tail also showed a dose-dependent increase in treated males. Mean body weight was decreased by 12% in MD males (6x the maximum clinical dose of 100 mg TID based on AUC_{0-6hr}) and by 23% and 9% in HD males (8x the maximum clinical dose of 100 mg TID based on AUC_{0-6hr}) and females (7x the maximum clinical dose of 100 mg TID based on AUC_{0-6hr}) respectively at the end of the treatment period. After the 4-week recovery period, mean body weight was still decreased by 10% in MD males and by 17% and 8% % in HD males and females respectively. Animals in the pair-fed control group also had decreased mean body weight (8%-10%) which was partially reversed to 6% at the end of the recovery period. Mean food consumption was decreased by 8% and 13% in MD and HD males and by 15% and 7% in the pair-fed control males and females respectively. At the end of the recovery period, mean food consumption showed partial recovery.

Sperm motility, sperm concentration and the number of normal sperms were decreased in all treated males relative to control. Amorphous sperms and sperms with decapitated heads were increased in all treated males relative to control. At the end of the treatment-free period, partial recovery of the altered sperm parameters was observed. It is likely full recovery will occur with time.

The target organs of toxicity include the GI tract – esophagus (serosal fibrosis), stomach (cytoplasmic vacuolation of chief cells), large intestine (mucosal necrosis), pancreas (acinal cell vacuolation), salivary gland (cytokaryomegaly – seromucus acinar cells), epididymides (hypospermia), testes (degeneration/atrophy), mesenteric lymph node (lymphoid depletion), skin of the tail (hyperkeratosis, acanthosis, pyogranulomatous dermatitis), eye (posterior synechia, cataract) and kidney (chronic progressive nephropathy). The incidence of testicular lesions was decreased at the end of the recovery period but the severity appear to have increased. The incidence of kidney lesions was decreased at the end of the recovery period. NOAEL could not be established because of the nephropathy and testicular lesions at the LD (3x the maximum clinical dose of 100 mg TID based on AUC_{0-6hr}).

STUDY TITLE: One-year oral toxicity study of SC-48334 in rats with a 4-week recovery period.

Key study findings:

- 7/60, 22/60, 23/60 and 18/60 animals were observed with palpable masses in the purina control, LD, MD and MHD groups, respectively. No dose-relationship was apparent in either the total number of animals affected (or the onset of occurrence at these dose levels. Only

2/60 animals in the HD group had palpable masses, however, this group was sacrificed early (during study week 20). In general, the masses were observed during the last month of treatment; masses were only observed in 3/60, 5/60, 12/60 and 6/60 animals in the control, LD, MD and MHD groups, respectively, during the last four weeks of dosing. Macroscopic and/or microscopic examination of the masses revealed them to be abscesses and/or associated with the mammary glands (lactation, galactoceles or active secretion). Only 3 masses were found to be tumors; an adenoma was present in 1/60 LD animal and 2/60 control group animals each had a fibroadenoma.

- Mortality occurred at all dose levels including controls. Due to the high mortality at HD, dosing was terminated during week 10 and sacrificed during week 20. 44/60 (HD), 15/60 (HMD), 9/60 (MH), 8/60 (LD), 3/60 (, — diet control) and 11/60 (, — diet control) animals were found dead or sacrificed moribund during SC-48334 administration. The cause of death or moribundity was not ascertained in 35/60 (HD), 7/60 (HMD), 2/60 (MD), 6/60 (LD), 3/60 (purified diet control) and 7/60 (, — diet control) animals. The causes of death established by sponsor include septicemia (1/60-HD, 1/60-MD); chronic renal disease (1/60-HD, 3/60-HMD, 2/60-MD); enteropathy (5/60-HD); gavage error (1/60-HD, 2/60-each for HMD, MD & LD); trauma (1/60-HD, 2/60-MD); pituitary tumor (1/60-HMD, 2/60-MD) and leukemia (1/60-, — diet control). Reviewer believes that demise of most animals was due to GI toxicity (stomach-ulcer, hyperkeratosis; cecum-mucosal necrosis, inflammation, hemorrhage colon-dilatation of crypts, necrosis, edema, inflammation; ileum and jejunum-villous atrophy), renal toxicity (nephrosis, nephropathy, vacuolation of tubular epithelium, inflammation, protein casts and mineralization of tubular epithelium and pelvis), hepatotoxicity (necrosis, cytoplasmic vacuolation, hemorrhage and lymphocytic infiltrate) and cardiac toxicity (cardiomyopathy, inflammation and necrosis of myocardial fiber) based on the histopathology data provided.
- A dose-dependent decrease in % mean body weight was observed in both treated males and females. At 420 mg/kg/d, the % decreases in mean body weight were 18% and 14% respectively for males and females. At 840 mg/kg/d, the values were 32% and 26% for males and females respectively.
- Diarrhea was observed at doses ≥ HMD. The incidence of diarrhea was very high especially in the HD group during the first 12 weeks of treatment (45/60 - weeks 1-4; 46/60 - weeks 5-8; 21/60 - weeks 9-12). Treatment was terminated in the HD group at week 10 due to mortality. However, the diarrhea persisted till week 12 and thereafter subsided completely. In the HMD animals, the incidence of diarrhea was 26/60 during the first four weeks of treatment but subsided significantly from week 5 onwards.
- Percent decrease in food consumption also showed a dose-dependent effect. At 420 mg/kg/d, the % decreases in food consumption were 13% and 5% respectively for males and females. At 840 mg/kg/d, the values were 26% and 9% for males and females respectively.
- Equatorial cataracts were observed in treated males relative to controls. The incidence appears to be dose-related. In treated males the incidence are1/28 (180mg/kg), 1/29 (420 mg/kg) and 18/27 (840 mg/kg) at week 52. In treated females, the incidence at the 840 mg/kg dose is 9/23 at week 52. After the 4 week recovery period, the incidence had decreased to are1/10 (180mg/kg), 1/9 (420 mg/kg) and 5/9 (840 mg/kg) in treated males and 4/8 for treated females. In the 1680 mg/kg dose group that was terminated at week 20, the incidence of cataracts was 9/9 (males) and 5/7 (females) at week 14.
- WBC increased in a dose-dependent manner achieving statistical significance in all treated males and in females dosed 420 and 840 mg/kg/d. RBC was slightly but statistically significantly decreased in males dosed 180 and 420 mg/kg/d and in females dosed 420 and 840 mg/kg/d. MCH was significantly increased in males dosed 420 mg/kg/d but decreased in females dosed 180 mg/kg/d. While MCHC was slightly but statistically significantly

decreased in all treated females, it was only decreased in males dosed 840 mg/kg/d. Platelets were dose-dependently and statistically significantly decreased in 420 and 840 mg/kg/d males and in the 840 mg/kg/d females. At the end of the recovery period, all affected hematology parameters had returned to normal limits.

- At week 52, K, P, BUN and Ca were slightly but statistically significantly increased in all treated males relative to control. K was slightly but statistically significantly increased in all treated females. P was dose-dependently and slightly but statistically significantly increased in females dosed 420 and 840 mg/kg/d. Creatinine and cholesterol levels were also significantly increased in males dosed 180 and 420 mg/kg/d. Mg was slightly but statistically significantly increased in the 180 mg/kg/d males. AST and ALT increased in a dose-dependent manner achieving statistical significance in males dosed 420 and 840 mg/kg/d and in females dosed 840 mg/kg/d. At the end of the recovery period, P and Na were slightly but statistically significantly increased in males and females dosed 840 mg/kg/d respectively.
- At the end of the treatment period (week 52), urine Ca was dose-dependently and statistically significantly increased in all treated males and in females dosed 420 and 840 mg/kg/d. Urine volume was statistically significantly increased in all treated females. Urine total protein was statistically significantly increased in males dosed 180 and 420 mg/kg/d but not in the 840 mg/kg/d group. All urinalysis parameters returned to normal limits at the end of the recovery period.
- Relative weight of the liver was slightly but statistically significantly increased in the180 and 420 mg/kg/d males and in females dosed 420 and 840 mg/kg/d. Since there is no correlative histopathology, the increment may be due to the increased AST and ALT levels. The weight changes in the liver were partially reversed in females and fully reversed in males at the end of the recovery period. Absolute weight of the epididymides decreased in a dose dependent manner achieving statistical significance in at the 420 and 840 mg/kg/d dose levels. At the end of the recovery period the 840 mg/kg/d males still had a significantly decreased epididymal weight. Weights (abs. and rel.) of the testes decreased in a dose-dependent manner achieving statistical significance at the 840 mg/kg/d dose level. The decreased weights persisted throughout recovery. The decreased relative weight of the testes correlates with the atrophy of the seminiferous tubules. Relative weight of the kidney was statistically significantly increased in females dosed 420 and 840 mg/kg/d. At the end of the recovery period, the relative kidney weight of females dosed 420 mg/kg/d was still significantly different from control. In males, while weights were not significantly different from controls after treatment, relative kidney weight was statistically significantly increased in males dosed 180 and 840 mg/kg/d at the end of the recovery period. This may be due to the hyperplasia, nephropathy and mineralization of the pelvis observed histologically at the end of the recovery period.
- The target organs of toxicity include the epididymides (hypospermia), kidney (nephropathy, lymphocyte infiltration, protein casts, hyperplasia and mineralization), GI tract (stomach-ulcer, hyperkeratosis; cecum-mucosal necrosis, inflammation, hemorrhage colon-dilatation of crypts, necrosis, edema, inflammation; ileum and jejunum-villous atrophy), testes (atrophy of seminiferous tubules, aspermatogenesis, edema, and hyperplasia of interstitial cells), mammary gland (galactocele, active secretion) and tail (suppurative inflammation). Most of these toxicities occurred at all dose levels and showed very little/no recovery at the end of the treatment free period.
- NOAEL could not be established since toxicities were observed at the LD tested.

Study no: PSA-91C-3490
Volume #, and page #: Vol. 3; pg. 955.

Conducting laboratory and location: ——————————

Date of study initiation: May 1989.
GLP compliance: Yes (USA, Japan).
QA report: yes (x) no ( )
Drug, lot #, radiolabel, and % purity: Lot #s 88K040-301I, 88K042-301I, 89K008-301J. No information on purity.
Formulation/vehicle: SC-48334 dissolved in distilled/deionized water.

Methods (*unique* aspects):
Dosing: Animals were dosed orally (gavage) with the test article at 180, 420, 840 and 1680 mg/kg/day divided into three equal doses administered TID at intervals of 8 hr apart. All dose groups received a diet of —— rodent chow except the Control group 2 and the 1680 mg/kg/d group that received purified diet (dextrose). The purified diet was used to minimize the test article-related diarrhea observed at higher dose levels in previous studies with SC-48334.
Species/strain: Rat/Crl:CD.BR
#/sex/group or time point (main study): 30/sex/group.
Satellite groups used for toxicokinetics or recovery: 20/sex/group used for TK.
Age: 5 weeks at study initiation.
Weight: 96 – 175 g (M); 90 – 148 g (F).
Doses in administered units: 180, 420, 840 and 1680 mg/kg/day.
Route, form, volume, and infusion rate: Oral (gavage), solution, 10 ml/kg.

Observations and times:
Clinical signs: Daily.
Body weights: Weekly.
Food consumption: Weekly.
Ophthalmoscopy: Conducted prior to study initiation on all animals and during weeks 9, 14, 26, 50 and 55.
EKG: Not evaluated.
Hematology: Blood samples were collected from animals fasted overnight. Blood samples were taken from 10 male and 10 female rats in control group 2 (purified diet) and all rats in the 1680 mg/kg/day toxicology group after 10 weeks of test article administration (reticulocyte and differential white blood cell counts only) and at the interim sacrifice (week 20), from 10 male and 10 female rats from each toxicology study group after approximately 16 and 32 weeks of test article administration, and from all animals at the end of dosing (week 52) and at the end of the recovery period (week 56).
Clinical chemistry: Blood samples were collected as indicated for hematology. Routine clinical chemistry was conducted.
Urinalysis: Urine samples were collected from the animals bled weeks 10, 16, 20, 32, 52 and 56. A 24-hour urine sample was collected from each animal using a metabolism cage. Urine samples were generally collected approximately one week prior to blood samples.
Gross pathology: Organs/tissues collected for gross pathology examination is indicated in the list of addendum.
Organs weighed: Organs weighed at the interim (week 20), terminal (week 52) and recovery (week 56) sacrifices are indicated in the list of addendum.
Histopathology: Tissues indicated in the list of addendum were examined microscopically from all animals in the control groups (—— and Purified diets) and in the 840 and 1680 mg/kg/day toxicology groups at the week 20 and/or 52 sacrifices, and all animals found dead or sacrificed moribund during the study. In addition, the epididymides, heart, kidneys, mammary glands,

testes and thymus from all animals in the 180 and 420 mg/kg/day toxicology groups at the week 52 sacrifice and all animals at the recovery (week 56) sacrifice were examined histologically.
Toxicokinetics: Blood samples for PK evaluation were collected from animals in the PK groups on the first day of dosing and during study weeks 10, 25 and 51 (weeks 11, 26 and 52 of dosing, respectively). On the first day of dosing, blood samples were collected from all animals (20/sex/group) at appropriate intervals at approximately 0.5, 1, 2, 4 and 8 hours after dosing). Due to the high mortality observed in the HD (1680 mg/kg/d) group, dosing was terminated during week 10 and all animals were sacrificed. Prior to sacrifice and following the last dose, blood samples were collected from all HD animals. Blood samples were collected during weeks 25 and 51 (week 26 and 51 of dosing) from 15 animals/sex in the 180, 420 and 840 mg/kg/day groups.

Results:
Mortality:

Dose (mg/kg/d) Males	**0 (diet)**	**0 (Purified diet)**	**180**	**420**	**840**	**1680**
Found Dead	1/30 (3)		1/30 (37)	1/30 (29)	1/30 (35)	1/30 (0)
Found Dead	1/30 (11)		1/30 (44)		1/30 (43)	2/30 (3)
Found Dead	1/30 (15)				1/30 (44)	1/30 (4)
Found Dead	2/30 (26)				1/30 (51)	3/30 (6)
Found Dead						3/30 (7)
Found Dead						1/30 (8)
Sacrificed Moribund						3/30 (3)
Sacrificed Moribund						3/30 (4)
Sacrificed Moribund						2/30 (7)
Sacrificed Moribund						1/30 (9)
Sacrificed Moribund						1/30 (10)
Total Deaths	**5/30**	**0/30**	**2/30**	**2/30**	**5/30**	**21/30***
Scheduled Sacrificed	15/25 (52)	30/30 (20)	18/28 (52)	19/29 (52)	16/26 (52)	9/9 (20)
Found Dead				1/10 (54)	1/10 (53)	
Recovery Sacrifice	10/10 (56)		10/10 (56)	9/9 (56)	9/9 (56)	
Dose (mg/kg/d) Females	**0 (diet)**	**0 (Purified diet)**	**180**	**420**	**840**	**1680**
Found Dead	1/30 (4)	1/30 (2)	1/30 (1)	1/30 (15)	1/30 (0)	2/30 (2)
Found Dead	2/30 (15)	2/30 (106)	1/30 (14)	1/30 (22)	2/30 (15)	2/30 (4)
Found Dead	1/30 (17)		3/30 (15)	1/30 (47)	1/30 (20)	1/30 (5)
Found Dead	1/30 (45)				1/30 (27)	1/30 (6)
Found Dead					1/30 (31)	1/30 (8)
Found Dead					2/30 (50)	3/30 (9)
Found Dead					2/30 (52)	1/30 (10)
Sacrificed Moribund	1/30 (135)		1/30 (39)	1/30 (38)		1/30 (3)
Sacrificed Moribund				1/30 (40)		3/30 (4)
Sacrificed Moribund				1/30 (46)		4/30 (5)
Sacrificed Moribund				1/30 (51)		2/30 (6)
Sacrificed Moribund						2/30 (7)
Total Deaths	**6/30**	**3/30**	**6/30**	**7/30**	**10/30**	**23/30***
Interim sacrifice						
Scheduled Sacrificed	15/25 (52)	28/28 (20)	15/24 (52)	15/23 (52)	12/20 (52)	7/7 (20)
Recovery Sacrifice	9/9 (56)		9/9 (56)	8/8 (56)	8/8 (56)	
M + F Deaths	**11/60**	**3/60**	**8/60**	**9/60**	**15/60**	**44/60**

*** Dosing stopped at week 10; Number in parenthesis indicate week of occurrence.**

Histopathology:

SUGGESTED CAUSE OF DEATH (BY SPONSOR)												
Gross Pathology of Animals Found Dead or Sacrificed Moribund												
DOSE (mg/kg/d)	0 diet		0 Purified diet		180		420		840		1680	
Sex	M	F	M	F	M	F	M	F	M	F	M	F
Animals/group	30	30	30	30	30	30	30	30	30	30	30	30
Animals found dead/sacrificed	5	6	0	2	2	6	2	7	5	10	21	23
Intubation error	2/5	3/6			1/2		1/2	1/7		2/10	1/21	1/23
Atrial thrombus							1/2					
Died during sinus blood collection										2/10		
Chronic renal dzs.								2/7	2/5	1/10		1/23
Septicemia								1/7				
Pituitary tumor								2/7		1/10		
Enteropathy												2/23
Mech. Trauma Vertebral column												1/23
Cause of death Undetermined	3/5	3/6		2/2	1/2	6/6		1/7	3/5	4/10	20/21	18/23

Clinical signs: Numerous clinical signs of toxicity were observed in the HD animals. The predominant signs consisted of atonia, unkempt appearance, hypothermia, red and swollen forepaws, a reddened distal end of the tail, redness of the anal orifice, a swollen lower lip of the mouth, puffiness of the extremities and facial area, exfoliation of the tail (medial to distal), apparent segmenting of the tail, scabbing on the nose, red, brown or yellow material and/or staining on the anogenital, urogenital and diarrhea. Although these findings were generally observed at more than one observation period, the majority of these findings were most prevalent one hour following the first daily dose. In addition, several findings (redness of the anal orifice, diarrhea and feces with red material) were generally observed more frequently in females than in males. Most of these findings were not observed following the termination of dosing (during week 10) or the frequency greatly decreased during the recovery period.

Similar findings judged to be related to treatment were noted at the lower dose levels of LD, MD and MHD. This included the following: the distal end of the tail was reddened in MD and MHD animals; the incidence of this finding was markedly decreased during the final three to four months of dosing. Exfoliation of the tail (medial to distal) and apparent segmenting of the tail occurred at the LD, MD and MHD levels. The frequency and onset of occurrence were dose-related, and the incidence of both findings generally decreased during the 4-week recovery period. The only other finding that appeared to be related to test article administration at the LD, MD and MHD was the apparent raised areas on the mid and proximal tail. The frequency of this finding was dose-related and greatly decreased during the recovery period.

Palpable ventral masses were noted in males and females in control group 1 and in the LD, MD, MHD and HD groups. During the study, more animals in the LD, MD and MHD groups had masses than in the control group. No dose-relationship was apparent in either the total number of animals affected (7/60, 22/60, 23/60 and 18/60 animals with masses in the control, LD, MD and MHD groups, respectively) or the onset of occurrence at these dose levels. Only 2/60 animals in the HD group had palpable masses, however, this group was sacrificed early (during study week 20). Masses were only observed in 3/60, 5/60, 12/60 and 6/60 animals in the control, LD, MD and MHD groups, respectively, during the last four weeks of dosing. Macroscopic and/or microscopic examination of the masses revealed them to be abscesses and/or associated with the mammary glands (lactation, galactoceles or active secretion). Only 3

masses were found to be tumors; an adenoma was present in 1/60 LD animal and 2/60 control group animals each had a fibroadenoma.

Incidence of Diarrhea: empty cells indicate zero incidence.

DOSE (mg/kg/d)	0 diet	0 Purified diet	180	420	840	1680
Weeks 1 - 4					26/60	45/60
Weeks 5 - 8					0/60	46/60
Weeks 9 – 12					0/60	21/60
Weeks 13-16					2/60	0/60
Weeks 17- 52					1/60	*
Weeks 53-56 (Recovery)					0/60	

* Treatment was terminated at week 10, sacrificed at week 20

Body weights: Males (g)

DOSE (mg/kg/d)	0 diet	0 Purified diet	180	420	840	1680
Pretest N	142 ± 15.4 30	141 ± 12.0 30	136 ± 20.4 30	136 ± 13.3 30	136 ± 13.6 30	140 ± 12.6 30
Week 20 N	544 ± 61.9 26	547 ± 58.1 30	521 ± 53.6 30	406 ± 40.8** 29	412 ± 68.1** 30	476 ± 27** 9
Week 52 N	663 ± 86.4 25	A	604 ± 74.8* 28	545 ± 44.2** 29	454 ± 67.1** 26	A
Week 56 Recovery: N	669 ± 44.3 10	A	592 ± 63.3* 10	577 ± 52.6* 9	459 ± 67.2** 9	A
% ↓ Mean B. wt.	**0**		**9**	**18**	**32**	

A = interim sacrifice at week 20; * p < 0.05; ** p < 0.01

Females (g)

DOSE (mg/kg/d)	0 diet	0 Purified diet	180	420	840	1680
Pretest N	129 ± 8.1 30	126 ± 8.0 30	124 ± 8.0 30	124 ± 9.9 30	125 ± 7.4 30	124 ± 9.3 30
Week 20 N	366 ± 48.6 25	348 ± 48.6 28	352 ± 44.7 25	354 ± 40.9 29	312 ± 32.9** 27	320 ± 29 7
Week 52 N	468 ± 92.2 24	A	429 ± 66.9 24	402 ± 40.5** 23	348 ± 34.7** 26	A
Week 56 Recovery: N	479 ± 83.3 9	A	438 ± 68.9 9	407 ± 53.3 8	409 ± 40.7 8	A
% ↓ Mean B. wt.	**0**		**8**	**14**	**26**	

% ↓ Mean B. wt. at week 52; A = interim sacrifice at week 20; * p < 0.05; ** p < 0.01

Food consumption: Males (g/animal/day)

DOSE (mg/kg/d)	0 diet	0 Purified diet	180	420	840	1680
Pretest N	21 ± 2.3 30	19 ± 2.0** 30	20 ± 2.2 30	19.8 ± 1.7 30	19 ± 2.1** 30	16 ± 4.9** 29
Week 20 N	27 ± 2.9 26	27 ± 3.3 30	28 ± 3.1 30	24 ± 2.7 29	25 ± 3.7 30	27 ± 3.3 9
Week 52 N	23 ± 4.3 25	A	21 ± 2.9 28	20 ± 4.1 29	17 ± 4.61** 26	A
Week 56 Recovery: N	24 ± 3.5 10	A	24 ± 2.5 10	24 ± 2.7 9	22 ± 4.2 9	A
% ↓ Fd. Con.	**0**		**9**	**13**	**26**	

% ↓ Fd. Con. At week 52; A = interim sacrifice at week 20; * p < 0.05; ** p < 0.01

Females (g/animal/day)

DOSE (mg/kg/d)	0 diet	0 Purified diet	180	420	840	1680
Pretest N	20 ± 1.7 30	20 ± 3.0 29	19 ± 2.2 30	19 ± 1.8 30	18 ± 1.9 29	16 ± 4.6** 30
Week 20 N	22 ± 3.2 25	25 ± 4.4* 28	23 ± 3.1 25	25 ± 3.2* 29	21 ± 2.8 27	27 ± 6.1* 7
Week 52 N	22 ± 3.5 24	A	22 ± 2.7 24	21 ± 2.7 23	20 ± 2.7 22	A
Week 56 Recovery: N	22 ± 3.8 9	A	22 ± 3.3 9	22 ± 2.6 8	24 ± 3.7 8	A
% ↓ Fd. Con.	0		0	5	9	

% ↓ Fd. Con. At week 52; A = interim sacrifice at week 20; * p < 0.05; ** p < 0.01

Ophthalmoscopy:

Males (equatorial cataracts/animals examined)

DOSE (mg/kg/d)	0 diet	0 Purified diet	180	420	840	1680
Week 9	0/29	0/30	1/30	1/30	16/30	7/10
Week 14	0/28	0/30	0/30	1/30*	18/30	9/9
Week 26	0/26		0/30	2/30*	24/30	
Week 52	1/25		1/28*	1/29**	18/27	
Week 55:recovery	0/10		1/10*	1/9**	5/9	

* or ** = same animal

Females (equatorial cataracts/animals examined)

DOSE (mg/kg/d)	0 diet	0 Purified diet	180	420	840	1680
Week 9	0/29	0/29	0/29	0/30	1/29	2/8
Week 14	0/29	0/29	0/29	3/30	12/28	5/7
Week 26	0/25		0/25	3/28	14/26	
Week 52	0/24		0/24	0/24	9/23	
Week 55:recovery	0/9		0/9	0/8	4/8	

Electrocardiography: No data.

Hematology:

Males - Week 52 data						
DOSE (mg/kg/d)	0 diet	0 Purified diet	180	420	840	1680
WBC (10^3/UL) N	10.6 ± 2.3 24	A	16.0 ± 4.9* 26	17.9 ± 6.2** 28	19.3 ± 9.0** 25	A
RBC (10^6/UL)	8.5 ± 0.4	A	8.0 ± 0.7*	7.9 ± 0.8**	8.3 ± 0.7	A
MCV (U^3)	55.0 ± 2.1	A	56.6 ± 2.2	57.5 ± 2.8**	56.7 ± 3.01	A
MCHC (%)	34.7 ± 0.8	A	34.6 ± 1.1	34.3 ± 1.1	33.5 ± 1.2**	A
PLT (10^3/UL)	1123 ± 188	A	1146 ± 182	852 ± 162**	782 ± 212**	A
Females - Week 52 data						
DOSE (mg/kg/d)	0 diet	0 Purified diet	180	420	840	1680
WBC (10^3/UL) N	8.0 ± 3.0 23	A	10.0 ± 2.1 22	11.9 ± 3.5** 22	13.0 ± 3.1** 22	A
HGB (g/dl)	15.6 ± 1.0	A	15.2 ± 0.68	14.7 ± 0.6**	15.0 ± 0.6*	A
MCH (pg)	21.6 ± 1.0	A	20.8 ± 0.8*	21.1 ± 1.1	20.5 ± 0.93	A
MCHC (%)	36.8 ± 1.36	A	35.0 ± 0.75**	35.2 ± 1.1**	34.3 ± 1.2**	A
PLT (10^3/UL)	1023 ± 118	A	1090 ± 136	933 ± 180	828 ± 137**	A

A = interim sacrifice at week 20; * p < 0.05; ** p < 0.01

By the end of the recovery period (week 56), all affected hematology parameters had returned to normal limits.

Clinical chemistry:

Males - Week 16 data						
DOSE (mg/kg/d)	**0 diet**	**0 Purified diet**	**180**	**420**	**840**	**1680**
AST (U/L)	125 ± 10.3	121 ± 15.1	141 ± 40.3	195 ± 90.7*	299 ± 99.8**	128 ± 25.7
Phosphorus (mg/dl)	9.1 ± 1.1	8.0 ± 1.4	10.1 ± 1.5	11.6 ± 2.5**	10.4 ± 1.3	8.2 ± 1.5
T Bilirubin (mg/dl)	0.2 ± 0.05	0.2 ± 0.05	0.2 ± 0.05	0.3 ± 0.05	0.3 ± 0.08**	0.3 ± 0.1
Males - Week 52 data						
AST (U/L)	156 ± 51.6	A	158 ± 56.9	227 ± 98.1**	352 ± 89.7**	A
ALT (U/L)	74 ± 23.6	A	69 ± 17.2	106 ± 44.1**	137 ± 37.5**	A
ALKP (U/L)	82 ± 18.4	A	46 ± 15.2**	65 ± 22.2**	62 ± 18.1**	A
Phosphorus (mg/dl)	7.2 ± 1.0	A	9.1 ± 2.6*	9.2 ± 2.8**	9.9 ± 2.1**	A
BUN (mg/dl)	12.2 ± 1.7	A	15.0 ± 4.0*	16.3 ± 4.6**	15.6 ± 2.1**	A
Creatinine (mg/dl)	0.6 ± 0.07	A	0.8 ± 0.15**	0.7 ± 0.14**	0.7 ± 0.10	A
Cholesterol (mg/dl)	74 ± 21.7	A	110 ± 43**	107 ± 44*	91 ± 42.3	A
Calcium (mg/dl)	11.3 ± 0.88	A	12.1 ± 0.90**	12.1 ± 0.97**	12.3 ± 0.93**	A
Potassium (mEq/l)	5.91 ± 0.6	A	6.94 ± 0.7**	6.87 ± 0.93**	7.52 ± 1.1**	A
Males - Recovery Week 56						
Phosphorus (mg/dl)	7.2 ± 0.65	A	7.9 ± 1.14	8.2 ± 1.02	8.5 ± 0.68*	A
Females - Week 16 data						
DOSE (mg/kg/d)	**0 diet**	**0 Purified diet**	**180**	**420**	**840**	**1680**
AST (U/L)	130 ± 19.1	107 ± 16.6	138 ± 18.2	143 ± 18.7	251 ± 60.3**	102 ± 12.6
ALT (U/L)	83 ± 21.1	49 ± 8.9**	59 ± 5.7**	58 ± 9.5**	78 ± 12.1	45 ± 5.4**
T Protein	7.5 ± 0.9	7.0 ± 0.4	7.1 ± 0.6	7.1 ± 0.6	6.5 ± 0.5**	6.6 ± 6.3*
Albumin	4.7 ± 0.5	4.2 ± 0.3	4.5 ± 0.5	4.3 ± 0.4	3.9 ± 0.5**	3.9 ± 0.3**
Females - Week 52 data						
AST (U/L)	149 ± 39.8	A	133 ± 36.7	145 ± 24.3	265 ± 148**	A
Phosphorus (mg/dl)	6.9 ± 1.2	A	7.9 ± 1.7	8.1 ± 1.3*	8.4 ± 1.5**	A
Potassium (mEq/l)	5.21 ± 0.62	A	5.93 ± 0.93*	5.85 ± 0.82*	6.04 ± 0.98**	A
Females - Recovery Week 56						
Sodium (mEq/l)	143 ± 1.2	A	144 ± 1.5	144 ± 1.2	145 ± 1.8*	A

A = interim sacrifice at week 20; * p < 0.05; ** p < 0.01

Urinalysis:

Males - Week 15 data						
DOSE (mg/kg/d)	**0 diet**	**0 Purified diet**	**180**	**420**	**840**	**1680**
T. Volume (ml)	24.1 ± 6.3	16.8 ± 8.5	37.8 ± 14.1**	38.3 ± 7.01**	38.7 ± 9.3**	15.4 ± 4.9
Urine Ca (mg/dl)	5.1 ± 1.7	3.3 ± 1.62	9.4 ± 5.1	18.1 ± 9.4**	27.2 ± 10.3**	3.9 ± 1.11
Males - Week 51 data						
Urine Ca (mg/dl)	5.2 ± 2.42	A	13.3 ± 5.40**	19.7 ± 6.24**	23.7 ± 7.48**	A
Urine T. protein (mg/dl)	148 ± 160	A	656 ± 515**	438 ± 346*	285 ± 337	A
Males - Recovery Week 56						
All affected parameters returned to normal limits.						
Females - Week 15 data						
DOSE (mg/kg/d)	**0 diet**	**0 Purified diet**	**180**	**420**	**840**	**1680**
T. Volume (ml)	21.3 ± 5.12	19.2 ± 10.0	30.7 ± 11.3	32.3 ± 16.0	36.4 ± 11.4*	20.4 ± 7.3
Urine Ca (mg/dl)	9.9 ± 4.0	4.1 ± 3.3	22.5 ± 9.24**	29.7 ± 8.27**	32.7 ± 8.14**	5.0 ± 2.22
Urine Cl (mEq/l)	110 ± 57.1	29 ± 13.1**	76 ± 29.1	70 ± 20.3*	78 ± 25.3	36 ± 10.3**
Urine Na (mEq/l)	56 ± 16.4	29 ± 9.5**	43 ± 17.1	39 ± 8.9*	35 ± 15.7*	27 ± 9.0**
Urine K (mEq/l)	155 ± 48.7	66 ± 26.2**	109 ± 41.1*	103 ± 32.5**	93 ± 22.8**	67 ± 15.0**
Females - Week 52 data						
T. Volume (ml)	34.2 ± 10.6	A	46.3 ± 14.3*	53.0 ± 20.9**	48.8 ± 15.3*	A
Urine Ca (mg/dl)	17.0 ± 5.9	A	21.3 ± 7.5	26.7 ± 10.4**	28.9 ± 8.9**	A

Females - Recovery Week 56
All affected parameters returned to normal limits.

A = interim sacrifice at week 20; * p < 0.05; ** p < 0.01

Organ weights: Absolute organ weights. Only animals in the control (purified diet) and HD groups were sacrificed at week 20. Relative weights = organ wt. relative to final body wt. (g/100 g)

Males - Week 20 data						
DOSE (mg/kg/d)	**0 diet**	**0 Purified diet**	**180**	**420**	**840**	**1680**
Kidney (g)		3.44 ± 0.36				2.9 ± 0.18**
Liver (g)		15.8 ± 3.09				11.4 ± 0.87**
Epididymides (g)		1.41 ± 0.16				0.69 ± 0.10**
Epididymides (g/100 g)		0.27 ± 0.026				0.16 ± 0.027**
Testes (g)		3.3 ± 0.52				1.2 ± 0.33**
Testes (g/100 g))		0.62 ± 0.12				0.26 ± 0.08**
Brain (g/100 g)		0.41 ± 0.05				0.46 ± 0.03**
Spleen (g/100 g)		0.15 ± 0.026				0.18 ± 0.26*
Pituitary gl. (g/100 g)		0.005 ± 0.001				0.006 ± 0.002*

Males - Week 52 data						
Heart (g)	1.88 ± 0.19	A	1.77 ± 0.26	1.71 ± 0.26	1.53 ± 0.20**	A
Heart (g/100 G)	0.29 ± 0.04	A	0.30 ± 0.05	0.32 ± 0.04	0.34 ± 0.05*	A
Liver (g)	19.8 ± 3.65	A	21.9 ± 3.97	19.2 ± 3.12	16.1 ± 3.33*	A
Liver (g/100 g)	3.06 ± 0.32	A	3.64 ± 0.47**	3.62 ± 0.60**	3.47 ± 0.41	A
Salivary glands (g)	0.86 ± 0.11	A	0.87 ± 0.12	0.74 ± 0.11**	0.64 ± 0.08**	A
Spleen (g)	0.97 ± 0.20	A	0.94 ± 0.16	0.83 ± 0.16	0.74 ± 0.15**	A
Epididymides (g)	1.42 ± 0.14	A	1.27 ± 0.16	1.03 ± 0.19**	0.86 ± 0.17**	A
Testes (g)	3.41 ± 0.37	A	3.30 ± 0.76	2.90 ± 0.89	1.49 ± 0.44**	A
Testes (g/100 g)	0.53 ± 0.07	A	0.56 ± 0.14	0.55 ± 0.16	0.33 ± 0.12**	A
Brain (g/100 g)	0.35 ± 0.05	A	0.38 ± 0.05	0.41 ± 0.04**	0.47 ± 0.06**	A
Adrenal gl. (g/100 g)	0.008 ± 0.0023	A	0.010 ± 0.0021	0.012 ± 0.0037**	0.012 ± 0.0026**	A
Pituitary gl. (g/100 g)	0.002 ± 0.0006	A	0.002 ± 0.0005	0.003 ± 0.0005	0.003 ± 0.0004**	A
Males - Recovery Week 56						
Salivary glands (g)	0.80 ± 0.067	A	0.82 ± 0.07	0.75 ± 0.01*	0.66 ± 0.11**	A
Brain (g/100 g)	0.34 ± 0.03	A	0.34 ± 0.05	0.38 ± 0.03	0.47 ± 0.08**	A
Kidneys (g)	4.06 ± 0.48	A	4.17 ± 0.50	3.89 ± 0.59	3.35 ± 0.56*	A
Kidneys (g/100 g)	0.62 ± 0.05	A	0.72 ± 0.09*	0.69 ± 0.07	0.75 ± 0.09**	A
Epididymides (g)	1.44 ± 0.17	A	1.20 ± 0.23	1.25 ± 0.24	0.83 ± 0.12**	A
Testes (g)	3.27 ± 0.94	A	3.21 ± 0.63	2.96 ± 0.97	1.11 ± 0.29**	A
Testes (g/100 g)	0.50 ± 0.13	A	0.57 ± 0.14	0.53 ± 0.18	0.25 ± 0.06**	A
Thyroid gland (g)	0.04 ± 0.004	A	0.03 ± 0.006	0.026 ± 0.005**	0.026 ± 0.006**	A
Heart (g/100 g)	0.27 ± 0.03	A	0.29 ± 0.03	0.29 ± 0.02	0.37 ± 0.10**	A
Adrenal gl. (g/100 g)	0.009 ± 0.002	A	0.010 ± 0.003	0.010 ± 0.002	0.014 ± 0.006**	A
Pituitary gl. (g/100 g)	0.002 ± 0.0008	A	0.003 ± 0.0005	0.002 ± 0.0003	0.003 ± 0.0007**	A

Females - Week 20 data						
DOSE (mg/kg/d)	**0 diet**	**0 Purified diet**	**180**	**420**	**840**	**1680**
Kidney (g)		2.67 ± 0.32				2.2 ± 0.22**
Liver (g)		12.2 ± 2.32				8.93 ± 0.91**
Liver (g/100 g)		3.58 ± 0.57				2.98 ± 0.24*

Females - Week 52 data						
Salivary glands (g)	0.66 ± 0.08	A	0.62 ± 0.06	0.63 ± 0.10	0.56 ± 0.07**	A
Ovaries (g)	0.12 ± 0.023	A	0.11 ± 0.022	0.11 ± 0.019	0.09 ± 0.017*	A
Brain (g/100 g)	0.46 ± 0.099	A	0.49 ± 0.090	0.53 ± 0.066	0.61 ± 0.032**	A

Heart (g/100 g)	0.33 ± 0.055	A	0.35 ± 0.053	0.37 ± 0.036	0.39 ± 0.028**	A
Kidney (g/100 g)	0.71 ± 0.11	A	0.82 ± 0.22	0.92 ± 0.22**	0.90 ± 0.10*	A
Liver (g/100 g)	3.35 ± 0.43	A	3.83 ± 0.72	4.12 ± 0.73**	4.03 ± 0.48**	A
Females - Recovery Week 56						
Heart (g/100 g)	0.31 ± 0.04	A	0.36 ± 0.07	0.39 ± 0.04*	0.40 ± 0.05**	A
Liver (g/100 g)	3.29 ± 0.44	A	3.91 ± 0.54	4.35 ± 0.71**	3.54 ± 0.18	A
Kidney (g/100 g)	0.72 ± 0.08	A	0.91 ± 0.24	1.08 ± 0.29**	0.84 ± 0.17	A
Uterus (g/100 g)	0.20 ± 0.07	A	0.31 ± 0.07*	0.27 ± 0.04	0.30 ± 0.09**	A

A = interim sacrifice at week 20; * p < 0.05; ** p < 0.01

Gross pathology: Animals found dead or sacrificed moribund at all dose groups were necropsied. Empty cells indicate zero incidence for a particular finding.

Gross Pathology of Animals Found Dead or Sacrificed Moribund												
DOSE (mg/kg/d)	**0 diet**		**0 Purified diet**		**180**		**420**		**840**		**1680**	
Sex	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**
Animals/group	30	30	30	30	30	30	30	30	30	30	30	30
Animals found dead/sacrificed	5	6	0	2	2	6	2	7	5	10	21	23
Adrenal glands Dark red	2/5			1/2				1/7	3/5	3/10	2/21	6/23
Adrenal glands Reddened	1/5	1/6						2/7				1/23
GI Tract Dark red contents	2/5	6/6			2/2	6/6		7/7	1/5	2/10	4/21	1/23
Stomach Dark red foci		1/6						3/7			6/21	6/23
Esophagus Hemorrhagic							1/2			1/10		
Esophagus Rupture										1/10		
Esophagus Perforation		2/6										
Heart Cardiomegaly							1/2		1/5			
Heart Atrial thrombus							1/2					
Kidney Dark red	2/5								1/5	1/10	2/21	2/23
Kidney Reddened cortico-medullary junction										1/10		1/23
Kidney Nephrosis								2/7	1/5			
Kidney White foci									1/5	1/10		1/23
Lungs White foamy contents	2/5	1/6		1/2						1/10		1/23
Lungs Mottled										1/10	2/21	
Lungs Dark red	1/5					2/6				1/10		
Lungs Clear fluid contents											1/21	
Pituitary Reddened						3/6		1/7		1/10	2/21	3/23
Pituitary Enlarged		3/6						2/7		1/10		
Testes												

Small									5/5			

Only animals in the control (purified diet) and HD groups were sacrificed at Week 20.

Gross Pathology of Animals Sacrificed At Week 20 (Interim sacrifice)												
DOSE (mg/kg/d)	**0 diet**		**0 Purified diet**		**180**		**420**		**840**		**1680**	
Sex	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**
Surviving Animals/group	26	25	30	28	30	25	29	29	30	26	9	7
# of Animals sacrif -iced at Week 20			30	28							9	7
Lymph node, Sub. Reddened			4/30	6/28							4/9	3/7
Lymph node, Sub. Enlarged			5/30	1/28								
Kidney Rough				3/28								1/7
Pituitary Enlatged				3/28								
Salivary glands Hemorrhagic area											1/9	
Spinal cord Red fluid beneath meninges											1/9	
Testes Small			1/30								9/9	
Thymus Hemorrhage			2/30									1/7
Stomach Dark red contents			1/30	1/30								1/30

Gross Pathology of Animals Sacrificed At Week 52 (Scheduled sacrifice)												
DOSE (mg/kg/d)	**0 diet**		**0 Purified diet**		**180**		**420**		**840**		**1680**	
Sex	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**
Surviving Animals/group	25	24	0	0	28	24	29	23	25	20	0	0
Animals sacrificed	15	15	0	0	18	15	19	15	16	12	0	0
Adrenal gland White foci		1/15				1/15		2/15		1/12		
Adrenal gland Red foci		2/15			1/18	3/15	1/19	4/15		2/12		
Heart White foci									2/16			
Heart Thickened						2/15						
Heart Red foci						1/15						
Kidney White foci								1/15		1/12		
Kidney Cysts	1/15				2/18	3/15	4/19	2/15	2/16	2/12		
Mammary gland Lactation						7/15		11/15		7/12		
Pituitary Enlarged		2/15				5/15		4/15		1/12		

Testes Small							4/19		7/16			
Testes Soft							3/19		12/16			
Thymus Hemorrhagic		1/15					2/19	2/15		1/12		
Uterus Red foci						1/15		1/15		1/12		
Uterus Cysts						1/15		3/15		1/12		
External surface Red matting		2/15			7/18	2/15	6/19	3/15	5/16	3/12		
Mass Subcutaneous		3/15			2/18	1/15		5/15	1/16			
Mass Submandibular								1/15				
Tail, raised areas Multifocal	1/15	2/15			6/18	2/15	8/19	10/15	9/16	6/12		
Tail Abscesses	2/15				1/18	1/15	1/19	1/15	1/16	2/12		
Lymph node Inguinal, enlarged							4/19	1/15				
Lymph node Inguinal, dark red						1/15		1/15				
Lymph node Inguinal, green							5/19			2/12		
Fat Necrosis							1/19	1/15				

Gross Pathology of Animals Sacrificed At Week 56 (Recovery sacrifice)												
DOSE (mg/kg/d)	**0 diet**		**0 Purified diet**		**180**		**420**		**840**		**1680**	
Sex	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**
Surviving Animals/group	10	9	0	0	9	9	9	8	9	8	0	0
Animals sacrificed	10	9	0	0	9	9	9	8	9	8	0	0
Lymph node, Sub Reddened		2/9			1/9	3/9	1/9	1/8	5/9	2/8		
Adrenal glands Red foci		1/9			1/9	2/9	1/9	1/8	1/9	2/8		
Heart White foci									1/9	1/8		
Heart Enlarged									1/9			
Ileum Dark red contents									1/9			
Kidneys Granular surface					1/9	3/9		3/8	2/9			
Kidney, pelvis Red streaks					1/9	1/9		2/8				
Mammary glands Lactation		2/9				5/9		6/8		3/8		
Ovaries Cysts						3/9		1/8		2/8		
Ovaries White foci		2/9				5/9		4/8		5/8		
Testes Small	1/9				1/9		2/9		9/9			
Mass Subcutaneous		1/9			1/9	3/9	1/9	3/8				
Tail												

Raised areas					2/9	2/9	2/9	2/8	2/9	3/8		
Fat Necrosis									1/9			

Histopathology: Incidence and severity.

Histopathology of Animals Found Dead or Sacrificed Moribund (**Males**)						
DOSE (mg/kg/d)	**0** **diet**	**0** **Purified diet**	**180**	**420**	**840**	**1680**
Animals/group	**30**	**30**	**30**	**30**	**30**	**30**
Animals found dead/sacrificed	**6**	**0**	**2**	**2**	**5**	**21**
Cecum Mucosal necrosis						1/21(2)
Cecum, crypts Suppurative inflammation						1/21(1)
Cecum Suppurative inflammation						2/21 1/21(2) 1/21(3)
Colon Dilatation of crypts						1/21(2)
Stomach Hyperkeratosis						4/21 3/21(2) 1/21(3)
Stomach Ulcer						2/21(2)
Epididymides Hypospermia			1/2(3)		5/5 1/5(3) 4/5(4)	4/21 1/21(3) 3/21(4)
Esophagus Hemorrhage				1/2(4)		
Heart Suppurative inflammation						1/21
Heart Cardiomyopathy	1/6(2)		1/2(3)		3/5 1/5(2) 2/5(3)	
Heart Atrial thrombus				1/2(4)		
Kidney, Vacuolization, cytoplasmic-tubular epithelium					1/5(3)	6/21(3)
Kidney, Mineralization Tubular epithelium-medulla						5/21 2/5(1) 1/5(2) 2/5(3)
Kidney, Protein casts Collecting tubules			1/2(2)	1/2(3)	3/5 2/5(2) 1/5(4)	1/21(1)
Kidney Nephrosis					1/5(3)	2/21(3)
Kidney Cystic tubules					1/5(2)	1/21(2)
Kidney Nephropathy	1/6(1)		1/2(2)	1/2(4)		
Kidney Mineralization, pelvis					2/5 1/5(1) 1/5(2)	
Kidney Suppurative inflammation					2/5 1/5(3) 1/5(4)	
Liver Vacuolization, cytoplasmic						2/21(2)
Liver Necrosis, hemorrhage			1/2(4)			1/21(2)

Liver Infiltrate, lymphocyte					1/5(2)	
Lungs Edema, alveolar	3/6(2)		1/2(2)	1/2(2)	1/5(2)	3/21 1/21(2) 2/21(3)
Lungs Suppurative inflammation	1/6(2)					6/21 1/21(1) 3/21(2) 2/21(3)
Lungs Hemorrhage, alveolar	1/6(2)					2/21(2)
Lungs Perivascular edema	2/6(2)				2/5(2)	
Lympg node, Mesenteric Lymphoid atrophy					1/5(3)	1/21(4)
Rectum Suppurative inflammation						2/21 1/21(1) 1/21(2)
Spleen Lymphoid atrophy					1/5(3)	6/21 1/21(2) 5/21(3)
Testes Aspermatogenesis			1/2(3)		5/5 1/5(3) 4/5(4)	14/21 1/21(2) 10/21(3) 3/21(4)
Testes Multinucleated giant cells			1/2(2)			8/21 4/21(2) 4/21(3)
Testes Atrophy, seminiferous tubules			1/2(2)		4/5 3/5(3) 1/5(4)	2/21 1/2(2) 1/2(3)
Thymus Lymphoid atrophy					2/5(3)	14/21 4/21(2) 9/21(3) 1/21(4)
Thyroid gland Cytoplasmic vacuolization						6/21 1/21(2) 4/21(3) 1/21(4)
Tail Suppurative inflammation				1/2(2)	3/5 2/5(2) 1/5(3)	4/21 3/21(2) 1/21(3)

Histopathology of Animals Found Dead or Sacrificed Moribund (Females)						
DOSE (mg/kg/d)	**0 diet**	**0 Purified diet**	**180**	**420**	**840**	**1680**
Animals/group	**30**	**30**	**30**	**30**	**30**	**30**
Animals found dead/sacrificed	**6**	**2**	**6**	**7**	**10**	**23**
Cecum Mucosal necrosis						1/23(2)
Cecum, crypts Suppurative inflammation						9/23 4/23(1) 5/23(2)
Cecum Hemorrhage						2/23 1/23(1) 1/23(2)
Cecum Vasculitis						2/23 1/23(2) 1/23(3)

Cecum Submucosal edema				1/7(3)		3/23 1/23(2) 2/23(3)
Cecum Suppurative inflammation						5/23 3/23(2) 1/23(3) 1/23(4)
Colon Dilatation of crypts						4/23 (2)
Colon Suppurative inflammation						9/23 1/23(1) 4/23(2) 3/23(3) 1/23(4)
Colon Necrosis						1/23(4)
Colon Submucosal edema						3/23 1/23(1) 2/23(2)
Stomach Hyperkeratosis						4/23(2)
Stomach Ulcer	1/6(3)			3/7 1/7(1) 2/7(2)		1/23(2)
Ileum Villus atrophy						2/23(2)
Jejunum Villus atrophy						2/23(2)
Esophagus Suppurative inflammation	2/6(4)				1/10(4)	1/23(4)
Heart Suppurative inflammation	1/6(2)			1/7(4)		2/23 1/23(1) 1/23(2)
Heart Cardiomyopathy	1/6(2)			1/7(2)	1/10(1)	1/23(2)
Heart Necrosis, myocardial fiber						1/23(2)
Kidney, Vacuolization cytoplasmic-tubular epithelium						4/23(2)
Kidney, Mineralization Tubular epithelium-medulla		1/2(3)		1/7(3)	2/10 1/10(1) 1/10(2)	17/23 2/23(1) 10/23(2) 3/23(3) 2/23(4)
Kidney, Protein casts Collecting tubules	1/6(2)		2/6(2)	1/7(2)	1/10(4)	
Kidney Nephrosis				1/7(2)		1/23(1)
Kidney Cystic tubules					3/10 2/10(2) 1/10(3)	5/23 2/23(1) 3/23(2)
Kidney Nephropathy	1/6(3)			3/7 2/7(1) 1/7(4)		
Kidney Mineralization, pelvis				2/7 1/7(2) 1/7(3)	6/10 5/10(2) 1/10(3)	
Kidney Suppurative inflammation				1/7(3)		

Liver Vacuolization, cytoplasmic	1/6(3)				1/10(1)	
Liver Necrosis, hemorrhage						2/23 1/23(2) 1/23(4)
Liver Infiltrate, lymphocyte					1/10(1)	1/23(1)
Lungs Edema, alveolar	1/6(3)					
Lungs Suppurative inflammation	1/6(2)			2/7 1/7(2) 1/7(4)		2/23(2)
Lungs Hemorrhage, alveolar	1/6(2)	1/2(2)	2/6 1/6(2) 1/6(3)		1/10(2)	1/23(2)
Lungs Perivascular edema	3/6 2/6(2) 1/6(3)	1/2(2)			2/10 1/10(2) 1/10(3)	1/23(3)
Lymph *node*, *Mesenteric* Suppurative inflammation						2/23 1/23(2) 1/23(3)
Rectum Suppurative inflammation						5/23 1/23(1) 2/23(2) 1/23(3) 1/23(4)
Spleen Lymphoid atrophy				1/7(3)		3/23 2/23(2) 1/23(3)
Thymus Lymphoid atrophy	2/6 1/6(3) 1/6(4)			4/7 3/7(3) 1/7(4)	1/10(3)	11/23 3/23(2) 6/23(3) 2/23(4)
Thyroid gland Cytoplasmic vacuolization						4/23 2/23(2) 2/23(3)
Tail Suppurative inflammation				1/7(3)	2/10 1/10(3) 1/10(4)	4/23 2/23(2) 2/23(3)

Please note that only surviving animals in the purified diet control and HD groups were sacrificed at week 20.

Histopathology of Animals Sacrificed at Week 20				
DOSE (mg/kg/d)	**0** **Purified diet**	**1680**	**0** **Purified diet**	**1680**
	MALES		**FEMALES**	
Animals/group	**30**	**30**	**30**	**30**
Animals sacrificed	**30**	**9**	**28**	**7**
Colon Dilatation of crypts		1/9(4)		
Colon Suppurative inflammation				1/7(1)
Colon Lymphoid hyperplasia				1/7(3)
Parathyroid Follicular hyperplasia				1/7(2)
Epididymides Hypospermia	1/30(4)	8/9(4)		
Heart Cardiomyopathy	3/30(1)	1/9(1)	1/28(2)	

Heart Cell infiltrate, lymphocyte		1/9(2)		
Kidney, Mineralization Tubular epithelium-medulla	3/30 1/30(1) 2/30(2)	7/9 1/9(1) 5/9(2) 1/9(3)	26/28 2/28(1) 16/28(2) 8/28(3)	6/7 2/7(1) 2/7(2) 2/7(3)
Kidney Cell infiltrate, lymphocyte	7/30(1)	1/9(2)	5/28 1/28(1) 4/28(2)	
Kidney Nephropathy	3/30 2/30(1) 1/30(2)		1/28(3)	
Kidney Hydronephrosis	1/30(2)			
Liver Necrosis	4/30(2)		7/28 3/28(1) 4/28(2)	1/7(2)
Liver Vacuolization	2/30 1/30(1) 1/30(2)	1/9(2)	1/28(1)	
Liver Cell infiltrate, lymphocyte	13/30 12/30(1) 1/30(2)	4/9 3/9(1) 1/9(2)	5/28(1)	4/7 2/7(1) 2/7(2)
Pituitary Adenoma, pars distalis			1/28P	
Pituitary Hyperplasia, pars distalis			1/28(2)	
Prostate Suppurative inflammation	19/30 3/30(1) 14/30(2) 2/30(3)	3/9 1/9(1) 2/9(2)		
Rectum Cell infiltrate, lymphocyte		1/9(3)		1/7(2)
Rectum Hemorrhage				1/7(1)
Stomach Suppurative inflammation	14/30 7/30(1) 7/30(2)	6/9 1/9(1) 5/9(2)	5/28 2/28(1) 3/28(2)	
Testes Aspermatogenesis	1/30(4)	8/9 4/9(3) 4/9(4)		
Testes Edema, interstitial		9/9 5/9(2) 4/9(3)		
Testes Atrophy, seminiferous tubules	1/30(3)	7/9(3)		

Please note that only surviving animals in the —— diet control, LD, MD and MHD groups were sacrificed at week 52. In addition, the epididymides, heart, kidneys, mammary glands, testes and thymus from all animals in the LD and MD groups were examined histologically at the week 52 sacrifice and the recovery (week 56) sacrifice.

Histopathology of Animals Sacrificed at Week 52				
DOSE (mg/kg/d)	**0 —— diet**	**840**	**0 —— diet**	**840**
	MALES		**FEMALES**	
Animals/group	**30**	**30**	**30**	**30**
Animals sacrificed	**15**	**16**	**15**	**12**

Lymph node, submandibular Lymphoid hyperplasia	1/15(2)	5/16 2/16(2) 3/16(3)	2/15 1/15(2) 1/15(3)	
Parathyroid Follicular hyperplasia				1/12(2)
Adrenal gland Hyperplasia, medilla		1/16(2)		
Adrenal gland Pheochromocytoma		1/16P		
Eye Retinal regeneration		1/16(4)		
Eye Suppurative inflammation		1/16(3)		
Lungs Granulomatous inflammation				1/12(1)
Lungs Cell infiltrate, lymphocyte		1/16(1)		
Pancreas Vacuolization, cytoplasmic		1/16(2)		
Pituitary gland Adenoma	1/15P		4/15P	
Pituitary gland Hyperplasia, pars distalis				1/12P
Stomach Dilatation, gastric glands	1/15(2)	4/16(2)		
Thyroid gland Vacuolization, cytoplasmic		2/16 1/16(2) 1/16(3)		
Thyroid gland Follicular hyperplasia		2/16(3)		
Urinary bladder Suppurative inflammation				2/12(2)
Urinary bladder Cell infiltrate, lymphocyte		1/16(3)		2/12(3)

For animals in the LD and MD groups, only the epididymides, heart, kidneys, mammary glands, testes and thymus from all animals were examined histologically at the week 52 sacrifice and the recovery (week 56) sacrifice.

Histopathology of Animals Sacrificed at week 52								
DOSE (mg/kg/d)	**0 diet**		**180**		**420**		**840**	
Sex	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**
Animals/group	**30**	**30**	**30**	**30**	**30**	**30**	**30**	**30**
Animals sacrificed	**15**	**15**	**18**	**15**	**19**	**15**	**16**	**12**
Epididymides Hypospermia			1/18(4)		3/19(4)		15/16 1/16(2) 14/16(4)	
Heart Cardiomyopathy	4/15 3/15(1) 1/15(2)	4/15 1/15(1) 3/15(2)	8/18 6/18(1) 2/18(2)	6/15 1/15(1) 515(2)	15/19 7/19(1) 5/19(2) 3/19(3)	12/15 8/15(1) 4/15(2)	11/16 1/16(1) 8/16(2) 2/16(3)	4/12 1/12(1) 3/12(2)
Kidney, Mineralization Tubular epithelium					1/19(4)	3/15 1/15(2) 2/15(3)	3/16 1/16(1) 2/16(2)	
Kidney Cell infiltrate, lymphocyte	3/15(1)	2/15(2)	12/18 1/18(1) 10/18(2) 1/18(3)	1/15(1)		4/15 2/15(2) 2/15(3)	5/16(2)	4/12 3/12(2) 1/12(3)

Kidney, Protein casts Collecting tubules	1/15(1)	5/15 4/15(2) 1/15(3)	8/18 1/18(1) 2/18(2) 4/18(3) 1/18(4)	12/15 2/15(1) 8/15(2) 2/15(3)	5/19 2/19(1) 3/19(2)	5/15 1/15(1) 2/15(2) 2/15(3)	7/16 1/16(1) 6/16(2)	7/12 5/12(2) 2/12(3)
Kidney Hyperplasia, pelvic epithelium	1/15(2)		2/18(2)	3/15 2/15(2) 1/15(3)	4/19 1/19(1) 3/19(2)	7/15 1/15(1) 4/15(2) 1/15(3) 1/15(4)	2/16 1/16(1) 1/16(2)	7/12 1/12(1) 5/12(2) 1/12(3)
Kidney Nephropathy	8/15 2/15(1) 6/15(2)	5/15 1/15(1) 3/15(2) 1/15(3)	16/18 1/18(1) 10/18(2) 5/18(3)	7/15 5/15(2) 2/15(3)	12/19 7/19(2) 2/19(3) 3/19(4)	5/15 1/15(1) 1/15(2) 2/15(3) 1/15(4)	12/16 10/16(2) 1/16(3) 1/16(4)	
Kidney Mineralization, pelvis		4/15(2)	2/18 1/18(2) 1/18(3)	4/15 1/15(1) 2/15(2) 1/15(3)	11/19 3/19(2) 8/19(3)	9/15 3/15(1) 5/15(2) 1/15(3)	8/16 1/16(1) 5/16(2) 2/16(3)	7/12 2/12(1) 5/12(2)
Kidney Suppurative inflammation			2/18(2)	1/15(1)			2/16(2)	1/12(2)
Mammary gland Galactocele		2/15(2)		5/15 1/15(2) 4/15(3)		8/15 2/15(2) 6/15(3)		2/12P
Mammary gland Active secretion				3/15(3)		4/15 3/15(2) 1/15(3)		8/12P
Testes Aspermatogenesis			2/18(4)		4/19 1/19(3) 3/19(4)		14/16 3/16(3) 11/16(4)	
Testes Edema, interstitial	2/15(2)		13/18 3/18(1) 9/18(2) 1/18(4)		11/19 4/19(1) 5/19(2) 2/19(3)		12/16 2/16(2) 9/16(3) 1/16(4)	
Testes Atrophy, seminiferous tubules			3/18 2/18(1) 1/18(3)		4/19(3)		15/16 2/16(2) 12/16(3) 1/16(4)	
Testes Hyperplasia, interstitial cell			2/18(1)		1/19(1)			
Thymus Lymphoid atrophy							1/16(3)	
Thymus Hemorrhage		1/15(2)	4/18(1)	3/15 1/15(1) 2/15(2)	1/19(1)	4/15(2)		
Thymus Thymoma							1/16	
Tail Suppurative inflammation	1/15(3)	1/15(3)	3/18 1/18(2) 2/18(3)	2/15(3)	6/19 1/19(2) 3/19(3) 2/19(4)	9/15 3/15(2) 5/15(3) 1/15(4)	9/16 5/16(2) 4/16(3)	7/12 1/12(2) 5/12(3) 1/12(4)

Histopathology of Animals Sacrificed at week 56 (Recovery)								
DOSE (mg/kg/d)	**0 diet**		**180**		**420**		**840**	
Sex	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**
Animals/group	30	30	30	30	30	30	30	30
Animals sacrificed	10	9	10	9	9	8	9	8
Epididymides Hypospermia	1/10(4)		1/10(2)		3/9(4)		9/9(4)	
Heart Cardiomyopathy	6/10 2/10(1) 4/10(2)	4/9 2/9(1) 2/9(2)	7/10 2/10(1) 5/10(2)	3/9(1)	5/9 1/9(1) 3/9(2) 1/9(3)	3/8 1/8(1) 2/8(2)	9/9 7/9(1) 1/9(2) 1/9(3)	3/8(1)
Kidney, Mineralization Tubular epithelium, medulla		1/9(2)	1/10(2)	1/9(2)		1/8(2)	2/9 1/9(2) 1/9(3)	2/8(3)
Kidney, Hyperplasia Pelvic epithelium	2/10(1)			2/9(2)	1/9(2)	6/8 2/8(1) 3/8(2) 1/8(3)	6/9 5/9(2) 1/9(3)	4/8 3/8(2) 1/8(3)
Kidney Nephropathy	7/10 3/10(1) 4/10(2)	5/9 2/9(1) 3/9(2)	10/10 3/10(1) 4/10(2) 3/10(3)	5/9 1/9(2) 3/9(3) 1/9(4)	8/9 1/9(1) 6/9(2) 1/9(3)	6/8 1/8(1) 3/8(2) 2/8(4)	4/9 2/9(1) 1/9(2) 1/9(3)	2/8 1/8(2) 1/8(3)
Kidney, Mineralization Pelvis	1/10(2)			3/9(2)	3/9 1/9(1) 1/9(2) 1/9(3)	5/8 2/8(2) 3/8(3)	4/9(4)	6/8 4/8(2) 2/8(3)
Mammary gland Galactocele		2/9(3)		5/9(3)		4/8 1/8(2) 2/8(3) 1/8(4)		3/8(3)
Mammary gland Active secretion						5/8 4/8(2) 1/8(3)		2/8 1/8(2) 1/8(3)
Testes Aspermatogenesis					2/9(4)		9/9(4)	
Testes Edema, interstitial	2/10 1/10(2) 1/10(3)		7/10 3/10(1) 3/10(2) 1/10(3)		5/9 2/9(1) 2/9(2) 1/9(3)		9/9 7/9(2) 2/9(3)	
Testes Atrophy, seminiferous tubules	2/10 1/10(2) 1/10(3)		1/10(2)		3/9(3)		9/9 2/9(2) 7/9(3)	
Testes Hyperplasia, interstitial cell			2/10(2)		1/9(2)			
Tail Suppurative inflammation			2/10(3)	1/9(3)	2/9(2)		2/9 1/9(2) 1/9(3)	3/8 1/8(2) 2/8(3)

Toxicokinetics:

Dose (mg/kg/d)	C_{max} (µg/ml)				T_{max} (hr)				$AUC_{0\text{-}8hr}$ (µg.hr/ml)			
Males	**Day 1**	**Week 11**	**Week 26**	**Week 52**	**Day 1**	**Week 11**	**Week 26**	**Week 52**	**Day 1**	**Week 11**	**Week 26**	**Week 52**
180	3.81	-	3.03a	11.18	1.00	-	4.00a	1.00	16.39	-	b	40.21
420	5.98	-	16.50	16.44	0.50	-	0.50	1.00	30.89	-	62.85	76.75
840	6.39	-	20.23	28.70	0.50	-	1.00	1.00	39.39	-	86.30	84.65
1680	37.98	73.4	-	-	4.00	2.00	-	-	210.54	317.13	-	-

Females	Day 1	Week 11	Week 26	Week 52	Day 1	Week 11	Week 26	Week 52	Day 1	Week 11	Week 26	Week 52
180	4.60	-	2.47a	14.94	1.00	-	4.00a	0.50	16.02	-	b	32.76
420	7.33	-	23.80	14.83	1.00	-	0.50	1.00	33.55	-	64.75	56.14
840	13.50	-	32.50	45.03	0.50	-	1.00	1.00	58.40	-	96.77	99.62
1680	36.13	87.20	-	-	0.50	1.00	-	-	197.71		-	-

- = blood samples not collected.
a = mean concentration values were not available at early time intervals (0.5 – 2 hr) and values reported for T_{max} and C_{max} may not be accurate.
b = sample quantity was not sufficient for reanalysis and concentration value was not available for AUC calculation.
c = plasma concentrations were available only at 2 time-points and the AUC value was not used for data interpretation.

Historical Incidence (%) and Severity of Myocardial Degeneration and Fibrosis Complex (Cardiomyopathy) in SD Rats (Modified from Ref. 3 i.e. Appendix 1).

	Age (12 Months)					
	Male	Female	Male		Female	
	n =		%	Grade	%	Grade
myocardial degeneration[a]	50	50	44	1.9	9	1.8
myocarditis[a]	50	50	60	2.0	16	1.8
myocardial fibrosis[a]	50	50	31	2.2	11	1.8

[a] The term myocardial degeneration and fibrosis complex is a combination of these histologic lesions.

Historical Incidence (%) and Severity of Cardiomyopathy in SD Rats in Searle Study SA3490.

	Age (13.5 months)					
Sex	Male	Female	Male		Female	
	n =		%	Grade	%	Grade
SC-48334 dosages						
0 mg/kg	30	30	37	1.6	30	1.7
180 mg/kg	30	30	53	1.6	30	1.6
420 mg/kg	30	30	67	1.8	53	1.7
840 mg/kg	30	30	77	1.9	27	1.4

Summary of Study Findings:

In a 52-week chronic toxicity study in the rat with a 4 week recovery period, OGT 918 was administered by oral gavage (30/sex/group) at doses of 0, 180, 420, 840 and 1680 mg/kg/day (divided as three equal doses administered TID at 8 hr intervals). Due to high mortality in the 1680 mg/kg/day animals, dosing was terminated in this group during Week 10 and sacrificed during week 20. Clinical findings judged to be treatment-related at the lower dose levels consisted of tail findings in the 180, 420 and 840 mg/kg/day groups and soft stool in the 840 mg/kg/day group. Inhibition of bodyweight gain occurred in the 420, 840 and 1680 mg/kg/day groups throughout the treatment period. Inhibition of food consumption occurred in the 840 mg/kg/day males and 1680 mg/kg/day animals throughout the treatment period.

44/60 animals in the HD group (no AUC data at week 52) were found dead or sacrificed moribund during SC-48334 administration. Of the 44/60 animals found dead or sacrificed moribund, the cause of death or moribundity was not ascertained in 35/60 animals. Death of the remaining animals was attributed to septicemia, chronic renal disease, mechanical trauma, gavage error and enteropathy. 15/60 HMD (10x and 11x the maximum clinical dose of 100 mg TID based on $AUC_{0\text{-}6hr}$ for males and females respectively) animals were found dead or sacrificed moribund during the study. The cause of death was not ascertained for 7/60 HMD animals. Death of the remaining animals was attributed to chronic renal disease, gavage error, bleeding technique and pituitary tumor. 9/60 animals in the MD group (9x and 6x the maximum clinical dose of 100 mg TID based on $AUC_{0\text{-}6hr}$ for males and females respectively) were found dead or sacrificed moribund. The cause of death was not ascertained in 2/60 animals. Death of the remaining animals was attributed to gavage error, chronic renal disease, pituitary tumor and septicemia. 7/60 LD (5x and 4x the maximum clinical dose of 100 mg TID based on $AUC_{0\text{-}6hr}$ for males and females respectively) animals were found dead or sacrificed moribund during the study. The cause of death was not ascertained in 6/60 animals. Death of the remaining animal was attributed to gavage error. 3/60 purified diet control females were found dead during the study. They were all females and the cause of death was not ascertained. In the —— diet control group, 11/60 animals were found dead or sacrificed moribund. The cause of death was not ascertained in 5/60 animals. Death of the remaining animals was attributed to gavage error and leukemia. Reviewer believes that demise of most of these animals was due to GI toxicity (stomach-ulcer, hyperkeratosis; cecum-mucosal necrosis, inflammation, hemorrhage colon-dilatation of crypts, necrosis, edema, inflammation; ileum and jejunum-villous atrophy), renal toxicity (nephrosis, nephropathy, vacuolation of tubular epithelium, inflammation, protein casts and mineralization of tubular epithelium and pelvis), hepatotoxicity (necrosis, cytoplasmic vacuolation, hemorrhage and lymphocytic infiltrate) and cardiac toxicity (cardiomyopathy, inflammation and necrosis of myocardial fiber) based on the histopathology data provided. All or most of the animals found dead had undergone autolysis, This might have confounded the histopathologic evaluation.

A dose-dependent decrease in % mean body weight was observed in both treated males and females. At 420 mg/kg/d (MD), the % decreases in mean body weight were 18% and 14% respectively for males and females. At 840 mg/kg/d (HMD), the values were 32% and 26% for males and females respectively. Percent decrease in food consumption also showed a dose-dependent effect. At 420 mg/kg/d, the % decreases in food consumption were 13% and 5% respectively for males and females. At 840 mg/kg/d, the values were 26% and 9% for males and females respectively. The decreased food consumption correlates with the decreased body weight.

Palpable ventral masses were observed in 7/60, 22/60, 23/60 and 18/60 animals in the —— control, LD, MD and MHD groups, respectively. No dose-relationship was apparent in either the total number of animals affected or the onset of occurrence at these dose levels. Only 2/60 animals in the HD group had palpable masses, however, this group was sacrificed early (during study week 20). Most were associated with mammary gland galactoceles or abscesses. Only 3 masses were found to be tumors; an adenoma was present in 1/60 LD animal and 2/60 control group animals each had a fibroadenoma. The other masses were abscesses or were associated with the mammary gland (lactation, galactoceles or active secretions).

Test article-related changes in hematology parameters, serum chemistry parameters and urinalysis were observed at all dose levels. After the recovery, all hematology values, serum chemistry values and urinalysis values were comparable to those in the control group.

Equatorial cataracts were observed in treated males relative to controls. The incidence appears to be dose-related. In treated males the incidence are1/28 (LD), 1/29 (MD) and 18/27 (HMD) at week 52. In treated females, the incidence at the HMD dose is 9/23 at week 52. After the 4 week recovery period, the incidence had decreased to are1/10 (LD), 1/9 (MD) and 5/9 (840 mg/kg) in treated males and 4/8 for treated females. In the HD group that was terminated at week 20, the incidence of cataracts was 9/9 (males) and 5/7 (females) at week 14. The cataracts may have been induced by the pharmacologic activity of the drug. OGT 918 causes perturbations in lipid metabolism. Perturbations in lipid metabolism are associated with lipid peroxidation, which generates free radicals. Oxygen free radicals are important known causes of tissue damage including cataracts.

Gross and microscopic findings indicated that the testis, epididymides, GI tract, eyes, heart and kidney were the principal test-article affected organs. Soft and/or small testes were seen grossly at LD, MD, HMD and HD. Apparently treatment and dose-related increases in aspermatogenesis, interstitial edema, and atrophy of seminferous tubules were observed microscopically in the LD, MD, HMD and HD animals. The incidence and severity of the lesions were essentially unchanged 4 weeks after cessation of treatment. Cardiomyopathy and nephropathy were the principal pathologic changes seen in the heart and kidneys in control, LD, MD and HMD animals found dead or sacrificed during the course or at termination of the study. The incidence of the heart and kidney findings increased with dose. Sponsor stated that these changes were considered to be species- or age-related. Literature supporting sponsor's claim is summarized in appendix 2.

The target organs of toxicity include the epididymides (hypospermia), kidney (nephropathy, lymphocyte infiltration, protein casts, hyperplasia and mineralization), testes (atrophy of seminiferous tubules, aspermatogenesis, edema, and hyperplasia of interstitial cells), GI tract (stomach-ulcer, hyperkeratosis; cecum-mucosal necrosis, inflammation, hemorrhage colon-dilatation of crypts, necrosis, edema, inflammation; ileum and jejunum-villous atrophy), eyes (cataracts), mammary gland (galactocele, active secretion) and tail (suppurative inflammation). Most of these toxicities occurred at all dose levels and showed very little/no recovery at the end of the treatment free period. NOAEL could not be established since toxicities were observed at the LD tested.

Study title: 52-Week Gavage Toxicity Study with SC-49483 in Cynomolgus Monkeys with 8 weeks of recovery.

Key study findings:

KEY STUDY FINDINGS:

- 10 animals (5/24 – control; 1/12 – LD; 4/24 – HD) died or were sacrificed in a moribund condition during this study; however, none of the deaths or unscheduled sacrifices were considered to be related to the test material. 8 of the 10 unscheduled deaths were directly related to sequelae of the gastric catheterization, most commonly inflammation and infection at the skin or stomach implantation site and peritonitis. Of the remaining two animals in the control group, one had significant inflammation of the large intestine, while the cause of moribundity of the other was not determined.
- The test material produced dose-dependent increases in fecal changes (white feces, yellowish substance in feces and organ changes during treatment. At the end of the recovery period, the fecal changes were still present in addition to red feces.
- QRS interval was slightly but statistically significantly increased by 0.02 sec in HD males relative to control. At the end of the recovery period, this parameter was comparable to that of control.

- RBC, HGB and HCT were slightly but statistically significantly decreased in HD females relative to control. Reticulocytes were statistically significantly increased by 50% in HD females relative to control. Prothrombin time was slightly but statistically significantly increased in HD females relative to control. At the end of the recovery period, RBC was still slightly but statistically significantly decreased in HD females. Platelets were statistically significantly increased in HD males by 136% relative to control. Basophils were slightly but statistically significantly decreased in HD males relative to control.
- Creatinine was slightly but statistically significantly decreased in the HD group relative to control. Total protein and chloride levels were also slightly but statistically significantly decreased in HD males. At the end of the recovery period, albumin was slightly but statistically significantly decreased in HD males whereas phosphate and bile acids were statistically significantly increased in HD females by 138% and 267% respectively relative to control. The increased bile acids may be suggestive of impaired liver function and cholestasis.
- Urine phosphorus was statistically significantly decreased by 71% in HD males relative to control. At the end of the recovery period, urine chloride excretion and urine calcium were statistically significantly decreased by 33% and 58% respectively in HD males and females.
- Mean sperm concentrations were highest for males in the control group (7.73×10^8), and decreased for males given 750 and 2000 mg/kg/day (4.02×10^8 and 2.96×10^8, respectively). This corresponds to 48% and 62% decrease in the LD and HD males. The number of amorphous sperms was 1% in HD males compared to zero for control.
- T4 was statistically significantly decreased in LD females by 32% and in HD males and females by 41% and 37% respectively relative to control. At the end of the recovery period, T4 was still statistically significantly decreased in HD females.
- The target organs of toxicity include the GI tract – cecum, colon, stomach (pigmented macrophages, granulomatous inflammation), Liver (pigmented macrophages, vacuolation), Pancreas (↓ zymogen granule staining), adrenal gland (mineralization), thyroid gland (vacuolated macrophage), seminal vesicle (mineralization), lymph nodes – mesenteric & submandibular (granuloma, mineralized, pigmented macrophage), mammary gland (lymphocytic infiltrate), skin (acanthosis/hyperkeratosis), cervix (lymphocytic infiltrate), skeletal muscle (inflammation, necrosis/degeneration), brain (mineralization, necrosis), spinal cord (mineralization) and kidney (angiectasis-glomerulus).
- NOAEL could not be established because of lesions in the liver, pancreas, seminal vesicle, brain and skeletal muscle at the LD.
- At the end of the recovery period, the following lesions were observed mostly in the HD group – Pancreas (↓ zymogen granule staining), submandibular salivary gland (atrophy, mineralization), Stomach (inflammation, fibrosis), heart, seminal vesicles, thyroid & parathyroid (lymphohistiocytic infiltrate), adipose tissue (fibrosis, inflammation, pigmented macrophage), cervix (mucopurulent exudate), spleen (fibrosis), skin (acanthosis/hyperkeratosis, fibrosis, crusting, ulceration), brain (mineralization), spinal cord (demyelination, axonal swelling) and kidney (tubular pigment, membranoproliferative glumerulopathy).

Study no: SA 4078
Volume #, and page #: Vol. 35, pg. 1.
Conducting laboratory and location:

Date of study initiation: August 12, 1993.
GLP compliance: Yes (U. S. A. and U. K.)
QA- Report: Yes (X) No ()

Drug, lot #, radiolabel, and % purity: Drug lot # 93K003-B2A; radiolabeled SC-49483 LOT #s GDS-2448-146, GDS-2448-148, GDS-3768-45, GDS-3768-47, GDS-3768-64, GDS-3768-65. 100% pure.
Formulation/vehicle: A solution of SC-49483 in a carrier made up of 0.5% methylcellulose (w/v), and 0.1% polysorbate 80 (v/v) in reverse osmosis water.

Methods (unique aspects):
Dosing: SC-49483 was administered at total daily doses of 750 and 2,000 mg/kg/d for 52 weeks. The total daily dose was divided into 3 doses/day each separated by 8 hours. The drug was administered through a catheter surgically implanted into the lumen of the stomach through week 19. After week 19, the catheters were tied off and animals were dosed by oral gavage up to week 52. A surgical control group (did not undergo any surgical or dosing procedures) and a vehicle control group were included in the study.
Species/strain: Monkey/Cynomolgus.
#/sex/group or time point (main study): Control: 12 animals/sex/group; 750 mg/kg/day: 6 animals/sex/group; 2000 mg/kg/day: 12 animals/sex/group.
Satellite groups used for toxicokinetics or recovery: 12 animals/sex/group (0 mg/kg/d); 6 animals/sex/group (750 mg/kg/day); 12 animals/sex/group (2000 mg/kg/day) for TK. 6 animals/sex/control and HD group were designated as recovery animals for 8 weeks after the 52 week treatment period.
Age: Young adult
Weight: 2.5 to 7.0 kg.
Doses in administered units: 750, 2000 mg/kg/day.
Route, form, volume, and infusion rate: gavage via intragastric catheter; 10 ml/kg/dose.

Group	Dosage Level (mg/kg/day)[a]	Dose Concentration (mg/mL)	Number of Animals Male	Number of Animals Female
1 (Control)	0	0	12[b]	12[b]
2 (Low)[c]	750	25.0	6	6
3 (High)[c]	2000	66.7	12[b]	12[b]
4 (Instrument Control)[d]	NA	NA	4	4

a. The dose volume was 10 ml/kg/dose. When administering [^{14}C] SC-49483, the volume administered was adjusted to maintain the nominal dose of µci/kg. Each animal received the daily dosage divided into three doses/day. The control group received the carrier only.
b. Six animals/sex were designated as recovery animals and continued without treatment for at least 8 weeks after at least 52 weeks of treatment.
c. Three animals/sex/group in groups 2 and 3 received [^{14}C] SC-49483 once on Day 1 and once each during Weeks 26 and 52.
d. The animals in Group 4 were maintained in the jacket and tether system but did not undergo the surgical procedure nor were dosed with test material or carrier.

Observations and times:
Clinical signs: Twice daily.
Body weights: Recorded prior to treatment, on day 1 of treatment and weekly thereafter.
Food consumption: Daily.
Ophthalmoscopy: Was conducted before initiation of treatment; during Weeks 4, 8, 12, 26, and 52; and during Weeks 56 and 60 (recovery animals).
EKG: Conducted before initiation of treatment; during Weeks 13, 26, 39, and 52; and during Week 56 (recovery animals).
Hematology: Blood was collected from all animals. Animals were fasted for ~ 24 hours prior to blood collection. Blood was collected twice before initiation of treatment (pre- and post-surgery), during weeks 16, 32 and 52; and during weeks 56 and 60 (recovery animals).

Clinical chemistry: The blood collected for hematology evaluation was also used for routine clinical chemistry evaluation including hormone analysis.
Urinalysis: Urine samples were collected for approximately 24 hours for routine urinalysis evaluation.
Gross pathology: Organs/Tissues collected for gross pathology examination is indicated in the list of addendum.
Organs weighed: Organs weighed are indicated in the addendum list.
Histopathology: Tissues collected for histopathology examination is indicated in the list of addendum.
Toxicokinetics: Blood samples were collected on Day 1 and during Weeks 26 and 52 at approximately 0.5, 1, 3, 5, and 8 hours after the first daily dose. For [^{14}C] SC-49483, blood samples were collected at approximately 0.5, 1, 3, 5, 8, and 24 hours after the [^{14}C] SC-49483 dose. Urine and feces were collected for 168 hours after each radioactive dose at approximately 24-hour intervals. Urine was collected into containers surrounded by wet ice.
Other:
Sperm evaluation: At scheduled sacrifice, sperm was collected from the left epididymis from each male animal and evaluated for concentration, motility and morphology.
Hormone analysis: Serum was harvested from blood samples collected during weeks 16, 32 and 52; and during weeks 56 and 60 from recovery animals. TSH, T3, T4 and testosterone levels were measured.

Results:

Mortality:

MORTALITY						
	Vehicle Control		750 mg/kg/day		2000 mg/kg/day	
Sex	**M**	**F**	**M**	**F**	**M**	**F**
Incidence	2/12	3/12	1/6	0/6	2/12	2/12
Total	**5/24**		**1/12**		**4/24**	

Ten animals (5 males and 5 females) died or were sacrificed in a moribund condition during this study. However, none of the deaths or unscheduled sacrifices were considered to be related to the test material. 8 of the 10 unscheduled deaths were directly related to sequelae of the gastric catheterization, most commonly inflammation and infection at the skin or stomach implantation site and peritonitis. Of the remaining two animals in the control group, one had significant inflammation of the large intestine, while the cause of moribundity of the other was not determined.

Clinical signs: Incidence of clinical signs during weeks 1-53. Rectal body temperature and respiration rate were unremarkable during the treatment and recovery periods.

Dose (mg/kg/d)	0 (vehicle)		750		2000		0 (No surgery nor vehicle)	
Sex	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**
White feces		2/12	4/6	4/6	12/12	12/12		
Yellowish substance in feces		1/12	3/6	1/6	12/12	12/12		
Red skin (abdomen)	1/12			1/6		3/12	¼	
Red skin (hind limbs)				1/12	3/12	2/12		
Skin (broken-L hind limb)		1/12	2/6	2/6	4/12	2/12		
Red nasal discharge	6/12	4/12	3/6	1/6	8/12	6/12		
RECOVERY (WEEKS 53-61)								
Red feces						2/12		
White feces					4/12	4/12		
Yellowish substance in feces					4/12	4/12		

Empty cells indicate zero incidence

Body weights: No treatment-related effects on body weight. At the end of the treatment period, mean body weight of treated animals and those of controls were not statistically significantly different.
Food consumption: No treatment-related effects.
Ophthalmoscopy: Unremarkable.
Electrocardiography:

Dose (mg/kg/d) Week 52	0 (vehicle)		750		2000		0 (No surgery nor vehicle)	
Sex	M	F	M	F	M	F	M	F
Heart rate (beats/min)	167 18.9		175 29.5		160 27.6			
P-R interval (sec) M SD	0.08 0.000		0.08 0.000		0.09 0.019			
QRS interval (sec)	0.03 0.008		0.03 0.009		0.05* 0.013			
Q-T interval (sec)	0.21 0.030		0.21 0.027		0.22 0.034			
RECOVERY (WEEKS 56)								
At the end of the recovery period the prolonged QRS interval (HD males) had returned to normal limits.								

* p< 0.05

Hematology:

WEEK 52 DATA

Dose (mg/kg/d)	0 (vehicle)		750		2000		0 (No surgery nor vehicle)	
Sex	M	F	M	F	M	F	M	F
RBC (E6/UL)		6.09 ± 0.369		5.05 ± 0.513		5.29 ± 0.264*		6.25 ± 0.580
HGB (g/dl)		11.1 ± 0.71		10.6 ± 0.50		10.2 ± 0.81*		11.4 ± 0.73
HCT (%)		36.3 ± 2.17		34.8 ± 1.75		33.5 ± 2.31*		37.5 ± 2.47
RETIC (%)		0.2 ± 0.18		0.2 ± 0.13		0.4 ± 0.15*		0.2 ± 0.10
PT (sec)		10.9 0.29		10.6 ± 0.29		11.4 ± 0.43*		10.8 ± 0.26
RECOVERY (WEEKS 60)								
Dose (mg/kg/d)	0 (vehicle)		750		2000		0 (No surgery nor vehicle)	
Sex	M	F	M	F	M	F	M	F
PLT (E3/UL)	380 ± 77.5				518 ± 62.4*			
BASO (E3/UL)	0.1 ± 0.4				0.0 ± 0.0*			
RBC (E6/UL)		6.26 ± 0.434				5.40 ± 0.606*		

* p< 0.05

Clinical chemistry:

WEEK 52 DATA

Dose (mg/kg/d)	0 (vehicle)		750		2000		0 (No surgery nor vehicle)	
Sex	M	F	M	F	M	F	M	F
Creat (mg/dl)	1.1 ± 0.16	7.9±0.30	1.2 ± 0.15	7.8±0.49	1.0 ± 0.11*	7.3±0.51*		
T. Pro (g/dl)	8.0 ± 0.58	±	7.7 ± 0.43	±	7.4 ± 0.68*	±		
Chol (mg/dl)	136±14.4	±	103±17.4*	±	105±18.8*	±		
RECOVERY (WEEKS 60)								
Dose (mg/kg/d)	0 (vehicle)		750		2000		0 (No surgery nor vehicle)	
Sex	M	F	M	F	M	F	M	F
ALB (g/dl)	4.5±0.26				4.0±0.26*			
I PHOS (mg/dl)		5.5±0.75				7.6±0.84*		
Bile acids (umol/l)		3.0±2.0				8.0±2.6*		

* p< 0.05

Urinalysis:

URINE CHEMISTRY WEEK 52 DATA

Dose (mg/kg/d)	0 (vehicle)		750		2000		0 (No surgery nor vehicle)	
Sex	M	F	M	F	M	F	M	F
U PHOS (mg/dl)	21 ± 14		11 ± 11.5		6 ± 6.0*			
RECOVERY (WEEKS 60)								
Dose (mg/kg/d)	0 (vehicle)		750		2000		0 (No surgery nor vehicle)	
Sex	M	F	M	F	M	F	M	F
CL EXCR (mmol)	7.8 ± 1.29		±		5.2 ± 0.87*			
U Ca (mg/dl)		15.9±4.22		±		6.6±4.83*		

* p< 0.05

Organ weights: No statistically significant differences in absolute or relative organ weights at the terminal or recovery sacrifices.

Gross pathology:

WEEK 52 DATA (TERMINAL SACRIFICE)

Dose (mg/kg/d)	0 (vehicle)		750		2000		0 (No surgery nor vehicle)	
Sex	M	F	M	F	M	F	M	F
Lung Red foci		¼		1/6	1/6	1/6		
Adhesions	2/5	¼	1/5	1/6	4/6			
Stomach Adhesions	0/5	2/4	3/5	5/6	2/6	2/6		
Dark foci	1/5							
Thickened wall						1/6		
Cecum Dark foci			1/5		1/6			
Colon Dark foci						1/6		
Ovary Cyst(s)				1/6		1/6		
Small				1/6				
RECOVERY SACRIFICE (WEEKS 60)								
Dose (mg/kg/d)	0 (vehicle)		750		2000		0 (No surgery nor vehicle)	
Sex	M	F	M	F	M	F	M	F
Liver Red foci					¼			
Spleen Opaque capsule		1/5				2/4		

GROSS PATHOLOGY (UNSCHEDULED DEATHS)

Dose (mg/kg/d)	0 (vehicle)		750		2000	
Sex	M	F	M	F	M	F
N	2	3	1	0	2	2
Lung Red foci			1/1			
Diffusely light		1/3				
Dark foci		1/3				
Kidney Thickened capsule		1/3			½	
Dark foci	½					
Heart Red foci					½	
Liver						

Light foci		1/3				
Mottled		1/3				
Spleen Large		2/3				
Mottled		1/3				
Granular surface		1/3				
Stomach Red foci						½
Catheter site-stomach Thickened	½		1/1			2/2
Diffusely red	½		1/1			
Red foci						½
Submandibular L. node Large					½	
Peritoneum/cavity Adhesions		1/3			½	
Pleura/cavity Adhesions						
Thickened		1/3			½	

Sperm evaluation:

Dose (mg/kg/d)	0 (vehicle)	750	2000
Sperm concentration[a] (10^8)	7.73 ± 11.14	4.02 ± 5.41	2.96 ± 3.97
Sperm morphology Amorphous	0.0 ± 0.0	0.0 ± 0.4	1.0 ± 1.6

[a] = number of sperms/gram of caudal tissue

Hormone analysis:

WEEK 52 DATA

Dose (mg/kg/d)		0 (vehicle)		750		2000		0 (No surgery nor vehicle)	
Sex		M	F	M	F	M	F	M	F
Testosterone (ng/dl)		212 285	32.0 9.52	242 157	23.1 2.66	223 182	32.8 12.9		32.8 13.44
TSH (µU/ml)	M SD	3.2 0.57	3.4 0.81	2.7 0.23	3.2 0.80	3.0 0.92	2.90 0.30		2.9 0.55
T4 (µg/dl)	M SD	3.4 0.58	4.1 0.61	3.6 0.92	2.8* 0.56	2.0* 0.46	2.6* 0.61		4.2 0.34
T3 (ng/dl)		No data was provided for T3.							

WEEK 56 DATA

Dose (mg/kg/d)	Testosterone (ng/dl)		TSH (µU/ml)		T4		T3	
Sex	M	F	M	F	M	F	M	F
0 mg/kg/d	77.5 13.33	34.2 8.35	4.4 0.91	4.5 0.31	3.8 0.77	3.9 0.64	111 24.2	95.8 11.64
2000 mg/kg/d	203 227.2	41.5 6.36	4.3 1.24	3.8* 0.45	2.9 0.42	3.1 0.65	87.8 25.84	116 16.7

* p< 0.05

WEEK 60 DATA (RECOVERY)

Dose (mg/kg/d)	Testosterone (ng/dl)		TSH (µU/ml)		T4		T3	
Sex	M	F	M	F	M	F	M	F
0 mg/kg/d	126 73.3	30.7 9.57	4.1 0.77	4.1 0.38	3.9 1.05	3.50 0.42	140 24.9	122 23.6
2000 mg/kg/d	131 106.2	45.9 22.03	4.1 1.23	3.6 0.50	2.9 0.80	2.6* 0.68	111 28.0	110 32.7

* p< 0.05

Histopathology:

WEEK 52 DATA (TERMINAL SACRIFICE)

Dose (mg/kg/d)	0 (vehicle)		750		2000	
Sex	**M**	**F**	**M**	**F**	**M**	**F**
Cecum Pigmented macrophage					2/6 1/6(1) 1/6(2)	
Colon Pigmented macrophage					1/6(2)	
Liver Pigmented macrophage	1/5(1)				1/6(1)	2/6 1/6(1) 1/6(2)
Vacuolation			1/5(2)	1/6(2)	1/6(1)	
Pancreas ↓ Zymogen granule staining			5/5 4/5(1) 1/5(2)	5/6 4/6(1) 1/6(2)	6/6 4/6(1) 2/6(2)	6/6 4/6(1) 2/6(2)
Stomach Granulomatous inflammation				1/6(1)		1/6(1)
Adrenal gland Mineralization					1/6(2)	
Thyroid Vacuolated macrophage						1/6(1)
Cervix Lymphocytic infiltrate						1/6(1)
Seminal vesicle Mineralization			2/5 1/5(1) 1/5(2)		1/6(1)	
Mesenteric L. node Granuloma, mineralized					1/6(1)	
Pigmented macrophage						1/6(1)
Submandubular L. node Pigmented macrophage						1/6(1)
Mammary gland Lymphocytic infiltrate						3/6 2/6(1) 1/6(2)
Skin Acanthosis/hyperkeratosis						1/6(1)
Skeletal muscle Subacute inflammation				1/6(1)	2/6 1/6(1) 1/6(2)	1/6(2)
Necrosis/degeneration		1/4(1)	1/5(1)	2/6 1/6(1) 1/6(2)	2/6 1/6(1) 1/6(2)	1/6(1)
Brain Mineralization, vascular			1/5(1)		1/6(1)	
Mineralization			1/5(1)		2/6(1)	1/6(1)
Necrosis, white matter			1/5(1)			
Spinal cord Mineralization, vascular					1/6(1)	
Kidney Angiectasis, Glomerulus					1/6(1)	

RECOVERY SACRIFICE (WEEK 61)						
Pancreas ↓ Zymogen granule staining						3/4(1)
Submandibular salivary gland Atrophy					1/4(1)	
Mineralization					1/4(1)	1/4(1)

Stomach Chronic active inflammation					1/4(2)	
Fibrosis, serosal					1/4(2)	
Heart Lymphohistiocytic infiltrate					1/4(1)	
Parathyroid Lymphohistiocytic infiltrate					1/4(1)	
Thyroid Lymphohistiocytic infiltrate						1/4(1)
Adipose tissue Fobrosis						1/1(3)
Pigmented macrophage						1/1(2)
Chronic active inflammation						1/1(2)
Cervix Mucopurulent exudate						1/4(3)
Seminal vesicles Lymphohistiocytic infiltrate					1/4(1)	
Spleen Capsular fibrosis	1/5(1)	1/5(1)				2/4 1/4(2) 1/4(3)
Skin Fibrosis, subcutaneous					1/4(2)	
Acanthosis/hyperkeratosis					1/4(2)	
Superficial crusting					1/4(3)	
Ulceration					1/4(3)	
Brain Mineralization		1/5(1)			2/4(1)	
Spinal cord Demyelination/Axonal swelling						1/4(1)
Kidney Tubular pigment						1/4(1)
Membranoproliferative glumerulopathy						1/4(3)

1 = minimal, 2 = slight, 3 = moderate, 4 = marked, 5 = severe

UNSCHEDULED SACRIFICE

Dose (mg/kg/d)	0 (vehicle)		750		2000	
Sex	**M**	**F**	**M**	**F**	**M**	**F**
Cecum Inflammation, lymphohistiocytic		1/3(2)				
Inflammation, chronic active	1/2(3)					
Colon Inflammation, lymphohistiocytic		1/3(2)				
Inflammation, chronic active	1/2(3)					
Jejunum Dilatation, lymphatic vessel	1/2(2)					
Liver Lymphohistiocytic infiltrate	1/2(1)	2/3(1)	1/1(1)		2/2 1/2(1) 1/2(1)	1/2(1)
Vacuolation, centrilobular					1/2(1)	
Necrosis	1/2(2)					
Suppurative inflammation		1/3(4)				
Chronic active inflammation		1/3(4)				
Hepatocellular degeneration		1/3(2)				
Fibrosis		1/3(2)				
Pancreas Lymphohistiocytic infiltrate		2/3(1)				
Vacuolation		1/3(1)				
↓ zymogen granules			1/1(1)			
Parotid salivary gland Lymphohistiocytic infiltrate	1/2(1)	2/3 1/3(1)				

		1/3(2)				
Submandibular salivary gland						
Lymphohistiocytic infiltrate		1/3(2)				
Stomach						
Chronic active inflammation		1/3(3)				
Erosion, junction of esophagus & stomach		1/3(2)				
Heart						
Hemorrhage					1/2(1)	
Lymphohistiocytic infiltrate		1/3(1)				
Adrenal gland						
Extramedullary hematopoiesis		1/3(2)				
Thyroid gland						
Lymphohistiocytic infiltrate	1/2(1)					
Skin – catheter site						2/2
Inflammation, chronic active	1/2(3)	1/3(3)	1/1(4)		1/2(2)	1/2(2) 1/2(4)
Inflammation, granulomatous			1/1(1)		1/2(2)	
Fibrosis	1/2(2)	2/3 1/3(2) 1/3(3)			1/2(2)	1/2(3)
Hemorrhage			1/1(2)			1/2(2)
Bacterial colony		1/3P				2/2P
Acanthosis/hyperkeratosis		1/3(3)				
Stomach – catheter site						
Lymphohistiocytic infiltrate					1/2(1)	
Chronic active inflammation – submucosa/muscularis	1/2(3)	2/3(3)	1/1(3)		1/2(3)	2/2 1/2(3) 1/2(4)
Hemorrhage (submucosa)	1/2(2)		1/1(3)			
Foreign material	1/2(2)	2/3(3)	1/1(2)		1/2(1)	
Bacterial colony						2/2P
Hemorrhage (mucosa)						1/2(1)
Fibrosis		1/3(3)				
Pericardial sac						
Lymphohistiocytic infiltrate		1/3(2)				
Edema		1/3(3)				
Peritoneal cavity						
Inflammation, granulomatous	1/2(3)	1/3(4)			1/2(4)	
Pleural cavity						
Inflammation, granulomatous					1/2(5)	
Fibrosis					1/2(1)	
Vagina						
Lymphohistiocytic infiltrate		3/3(2)				2/2(1)
Axillary lymph node						
Lymphoreticular hyperplasia		1/3(4)				
Lumbar/iliac lymph node						
Lymphoreticular hyperplasia		1/3(4)				
Lymphocytic hyperplasia						1/2(2)
Hemorrhage						1/2(1)
Mesenteric lymph node						
Edema		1/3(2)				
Lymphoid depletion		1/3(2)				
Hemorrhage						1/2(1)
Germinal centers, decreased	1/2(2)					
Submandibular lymph node						
Hemorrhage						1/2(2)
Depletion, lymphoid		1/3(2)				
Renal lymph node						
Hemorrhage						1/2(2)
Mesentery						