11. Microscopic Pathology: Macro and microvesicular vacuoles were observed in the periportal locations of the liver in 4/10 males and 2/10 females in the high dose group.

12. Plasma Level of Test Drug: Mean plasma concentrations were 1.3, 9.1 and 104.8 ng/ml (males) or 1.4, 5.9 and 85.7 ng/ml (females) in the low, mid and high dose groups, respectively.

In summary, in the 26-week oral toxicity study in rats, SDZ HTF 919 was given to rats in feed at 0, 15, 60 and 240 mg/kg/day for 26 weeks. Major treatment related changes were mainly in the high dose group and these included hair loss, decreased terminal body weight gains (22.4-36%), food consumption (11-24%), monocytes (males, 37%) and reticulocytes (males, 28-30%), increased platelet count (males, 20-23%) and histopathological changes (macro and microvesicular vacuoles in the periportal locations of the liver). The no effect dose was identified at 60 mg/kg/day. The MTD was between 60 mg/kg/day and 240 mg/kg/day. The liver was the target organ of toxicity.

DOG:

2-week toxicity study with SDZ HTF 919 in dogs
(1 hour intravenous infusion)
338422)

Testing Laboratories:

Study Start and Completion Dates: January 22, 1993 and April 27, 1993

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Males (8.3-11.3 kg, 6.5-8.5 months)
Females (6.2-9.0 kg, 6.5-8.5 months)
Pure-bred beagle dogs

Methods: To evaluate the toxicity of HTF 919 in dogs, intravenous infusion (via cephalic veins) of HTF 919 was given to dogs for a period of 1 hour per day at 0 (0.9% NaCl), 0.1, 1.0 mg/kg/day, and a placebo control for 2-weeks. Clinical signs of

toxicity were observed twice daily. Mortality/viability were observed twice daily. Body weights were determined before the treatment and twice weekly thereafter. Food consumption was determined before the treatment and daily thereafter. Ophthalmoscopic examinations were conducted before the treatment and in week 2. ECGs were recorded before the treatment and on Day 1 and in week 2. Hematology, clinical chemistry and urinalysis were determined before the treatment and in week 2. All animals were necropsied at termination and the organs were weighed. The toxicokinetic profile was determined on day 1 after the treatment and at termination.

Results:

1. Clinical Signs: The adverse reaction to the treatment (swelling) at the infusion sites was observed in the 1.0 mg/kg group. Tremor, salivation and serous rhinorrhea were seen in all groups including control and placebo groups.

2. Mortality: There were no deaths.

3. Body Weight: Body weight gain was not markedly affected.

4. Food Consumption: Food consumption was not markedly affected.

5. Ophthalmoscopy: There were no treatment related alterations seen during the study.

6. ECG: There were no treatment related alterations seen during the study.

7. Hematology: There were no treatment related alterations seen during the study.

8. Clinical Chemistry: There were no treatment related alterations seen during the study.

9. Urinalysis: There were no treatment related changes observed during the study.

10. Organ Weights: Organ weights were not markedly affected.

11. Gross Pathology: Redness and/or thickness of the tissue at the infusion sites were seen in all groups.

12. Microscopic pathology: There were inflammation, hemorrhage, phlebitis and thrombus formation in the subcutis at the infusion sites in all groups including control and placebo groups but the changes in the high dose group were more severe.

13. Toxicokinetics: The plasma concentrations were proportional to the administered doses. C_{max} (352.4 ng/ml) in the high dose female group after the last dose was higher than that (225.5 ng/ml) after the first dose, suggesting that there were some drug accumulations over time. Following the administration of HTF 919 at 0.1 mg/kg/ day, C_{max} was ~1.5-1.9 times higher in the males than in the females. However, in the 1.0 mg/kg/day groups, C_{max} was ~2-3 folds higher in the females than in the males. The toxicokinetic measurements were summarized in the sponsor's table on page 8-1502 in Volume 1.6. This table is attached below.

PLASMA CONCENTRATIONS (ng/ml) AND PK PARAMETERS IN DOGS

SUMMARY OF RESULTS

	First application				Last application			
	0.1 mg/kg		1.0 mg/kg		0.1 mg/kg/day		1.0 mg/kg/day	
Time (h)	Males	Females	Males	Females	Males	Females	Males	Females
0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	3.4
1.0833	12.8	8.4	110.0	225.5	17.9	9.6	119.8	352.4
1.25	6.8	4.5	66.3	130.3	9.4	5.2	77.0	220.8
1.5	5.1	3.3	45.8	82.7	6.4	3.6	48.8	140.2
2	3.4	2.2	33.9	58.6	5.1	2.0	33.5	86.5
3	1.3	0.0	19.4	37.1	2.9	0.0	21.0	59.1
5	0.0	0.0	12.1	18.0	1.2	0.0	13.2	31.3
8	0.0	0.0	6.6	10.4	0.0	0.0	8.7	18.8
25	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
AUC (0-2h)	12.2	7.9	108.2	213.7	16.8	8.9	117.6	342.2
AUC (0-2h)/Dose	168.4	109.9	149.8	296.0	232.5	123.4	162.8	474.0
AUC (0-25h)			250.5	446.9			285.6	740.2
AUC (0-25h)/Dose			346.9	619.0			395.5	1025.3
Cmax (ng/ml)	12.8	8.4	110.0	225.5	17.9	9.6	119.8	352.4
CL (ml/min)							444.8	135.4

In summary, in the 2 week i.v. toxicity study in dogs, intravenous infusion of HTF 919 was given to dogs for a period of 1 hour per day at 0, 0.1 and 1.0 mg/kg/day for 2 weeks. The

adverse reaction to the treatment (swelling) was observed at the infusion sites in the 1.0 mg/kg group. Tremor, salivation and serous rhinorrhea were seen in all groups including control and placebo groups. Histopathological examination revealed that inflammation, hemorrhage, phlebitis and thrombus formation in the subcutis at the infusion sites in all groups including control and placebo groups but the changes in the high dose group were more severe.

26-week oral toxicity study in dogs
(SANDOZ 395D)

Testing Laboratories: Sponsor's laboratory,
[ ]

Study Start and Completion Dates: October 3, 1991 and May 7, 1993

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Males (8.4-11.5 kg, 7-7.5 months)
Females (7.5-9.8 kg, 7-7.5 months)
Beagle dogs

Methods: To evaluate the toxicity of HTF 919 in dogs, HTF 919 ——— was given orally to dogs at 5, 15 and 50 mg/kg/day for 26 weeks. The high dose (50 mg/kg/day) was increased to 60 mg/kg/day from week 14. Clinical signs of toxicity and mortality were observed daily. Body weights and food consumption were determined weekly. ECGs were recorded before treatment, in weeks 1, 13, 26 and 30. Ophthalmoscopic examinations were conducted before the treatment and in week 26. Hematology and clinical chemistry were determined before treatment and in weeks 6, 13, 26 and 30. Urinalysis was determined in week 26. All animals except the recovery animals were necropsied at termination (week 26) and the organs were weighed. The recovery animals were sacrificed in week 31. The toxicokinetic profile was determined on day 1 and in week 26.

Results:

1. Clinical Signs: The clinical signs including vomiting, diarrhea and hypersalivation were observed mainly in the high

dose group (50/60 mg/kg). Sporadic vomiting and diarrhea were also seen in the 5 and 15 mg/kg groups. Occasional hypersalivation was seen in two animals in the 15 mg/kg groups.

2. Mortality: There were no deaths.

3. Body Weight: Slight loss of body weight (up to ~8%) was observed in all treatment groups in the first two weeks. However, the total body weight gain at the end of the treatment period (26 weeks) was not markedly different between the control and treatment groups.

4. Food Consumption: Food consumption was significantly reduced (~19-61%) mainly in the first week in the all treated (male) groups and 15 and 50/60 mg/kg (female) groups.

5. Ophthalmoscopy: There were no treatment related alterations seen during the study.

6. ECGs: There were no treatment related alterations seen during the study.

7. Hematology: There were no treatment related changes observed during the study.

8. Clinical Chemistry: There were no treatment related changes observed during the study.

9. Urinalysis: There were no treatment related changes observed during the study.

10. Organ Weights: Organ weights were not markedly affected.

11. Gross Pathology: There were no treatment related changes observed during the study.

12. Microscopic Pathology: There were no treatment related changes observed during the study.

13. Toxicokinetics: Mean plasma concentrations were proportional to the administered doses. The mean maximum plasma concentrations measured after 26-week treatment were higher than those after the first treatment, suggesting that there was drug accumulated over time. The toxicokinetic measurements were summarized in the sponsor's table on page 8-887 in Volume 1.5. This table is attached on the following page.

Means ± standard deviations (rounded) of normalized Cmax and AUC's

Dose	5 mg/kg/day	15 mg/kg/day	60 mg/kg/day
	SINGLE APPLICATION		
Number of dogs	6	6	6
Cmax (ng/ml)	143 ± 92	317 ± 139	677 ± 435
Cmax/Dose (ng/ml for 1mg/kg)	40 ± 26	29 ± 13	19 ± 12
AUC/Dose (ng/ml.h for 1mg/kg)	131 ± 92	96 ± 57	92 ± 96
Dose	5 mg/kg/day	15 mg/kg/day	60 mg/kg/day
	26-WEEK TREATMENT		
Number of dogs	4	6	6
Cmax (ng/ml)	215 ± 30	524 ± 198	987 ± 335
Cmax/Dose (ng/ml for 1mg/kg)	60 ± 8	48 ± 18	23 ± 8
AUC/Dose (ng/ml.h for 1mg/kg)	221 ± 80	228 ± 131	218 ± 148

In summary, in the 26 week oral toxicity study in dogs, HTF 919 ——— was given orally to dogs at 5, 15 and 50/60 mg/kg/day for 26 weeks. Vomiting, diarrhea and hypersalivation were observed mainly in the high dose group (50/60 mg/kg). There were no treatment related pathological changes observed during the study. The gastrointestinal tract is the target organ of toxicity based on the clinical signs of toxicity (vomiting and diarrhea).

Addendum: There were two new 4-week oral dose ranging studies in dogs first time submitted to this NDA (study #88PS/D and #130 DF/D). These studies were not GLP studies. In study #88PS/D, dogs (1/group) were treated with HTF 919 orally ——— batch #906/502) at 3, 10, 30,and 100 mg/kg/day for 4 weeks (there was no control group). Major treatment related changes were observed in the high dose group and these included hypersalivation, hemorrhagic vomiting, reduced motor activity, body weight loss, dehydration, and histopathological changes (subacute gastritis). In the study #130DF/D, 1 male dog and 1 female dog were treated orally with HTF 919 (batch #91903) at 30 and 60 mg/kg/day,

respectively for 4 weeks. There were no clear treatment related changes.

A 52-week Oral ——— Toxicity Study in Dogs
(95/ — 019/0592)

Testing Laboratories: ———————————————
Sponsor's laboratory (toxicokinetic data)

Study Start and Completion Dates: March 4, 1994 and
August 14, 1997

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Males (9.5 ± 0.6 kg, 14 weeks)
Females (8.7 ± 0.7 kg, 14 weeks)
Beagle dogs

Batch no: 94904

Methods: To determine the potential toxicity of SDZ HTF 919 in dogs, SDZ HTF 919 ——— was given to dogs (4/sex/group) at 0, 5, 15 and 60/70 mg/kg/day for 52 weeks. The basis of dose selection was not provided. Due to lack of toxicity at 60 mg/kg/day, the dose was increased to 70 mg/kg/day in week 18. Clinical signs of toxicity and mortality were observed daily. Body weights and food consumption were determined weekly. Hematology, clinical chemistry and urinalysis were determined before the treatment, at weeks 5, 12, 25, 38 and 51 after the treatment. Ophthalmoscopy was performed before the treatment and at weeks 25 and 51 after the treatment. ECG and blood pressure were recorded before the treatment, at weeks 1, 25 and 51 after the treatment. All animals were necropsied at termination and the organs were weighed. Gross and microscopic examinations were then conducted. The toxicokinetic profile was determined before dosing, at 0.5, 1, 2, 4, 7 and 24 hours on the first day, at weeks 17, 26 and 52.

Results:

1. Clinical Signs: Salivation was seen mainly in the high dose group and occasionally in the low and mid dose groups.

2. Mortality: One high dose female was sacrificed during week 39. This dog had weight loss, low food consumption, salivation, hunched posture, body tremor and severe bacterial infection revealed by clinical chemistry and pathological examination.

3. Body Weight: The initial and final body weights in the control group were 9.5 ± 0.6 and 13.6 ± 0.7 (males) and 8.7 ± 0.7 and 11.8 ± 1.0 (females) kg, respectively. Terminal body weight gains were decreased by 27% (males) or 13% (females) in the high dose group. The body weight information is summarized in the following table.

Mean body weights (kg)

	0 mg/kg/day	5 mg/kg/day	15 mg/kg/day	60/70 mg/kg/day
Males				
Initial	9.5	9.3	9.0	9.5
26 weeks	13.2	12.3	12.2	12.2
52 weeks	13.6	13	12.8	12.5
Females				
Initial	8.7	9.2	8.8	8.7
26 weeks	11.2	12.0	11.1	10.8
52 weeks	11.8	13.0	11.8	11.4

The information was also depicted in figures 2A and 2B on pages 42 and 43 in volume 18.4. These figures are attached below.

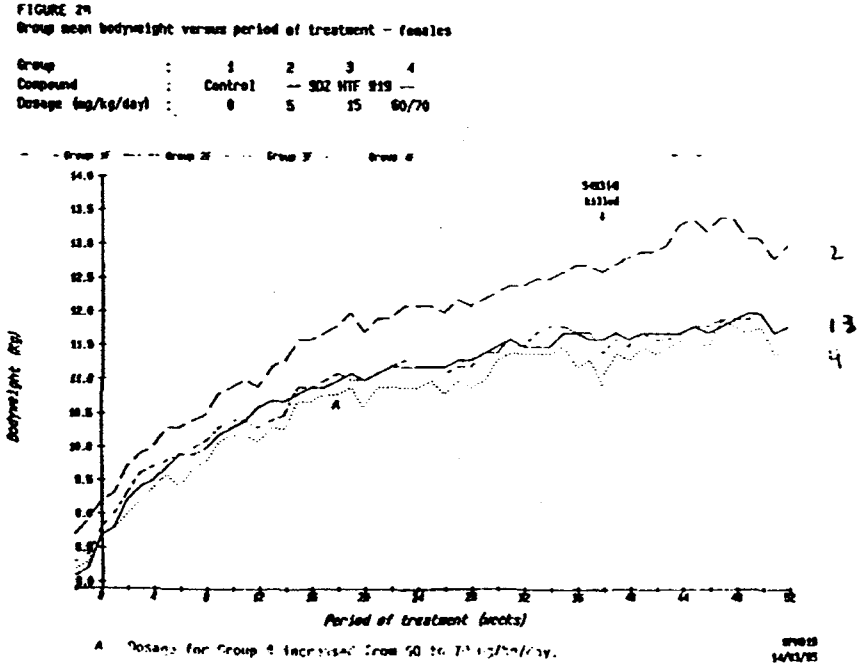
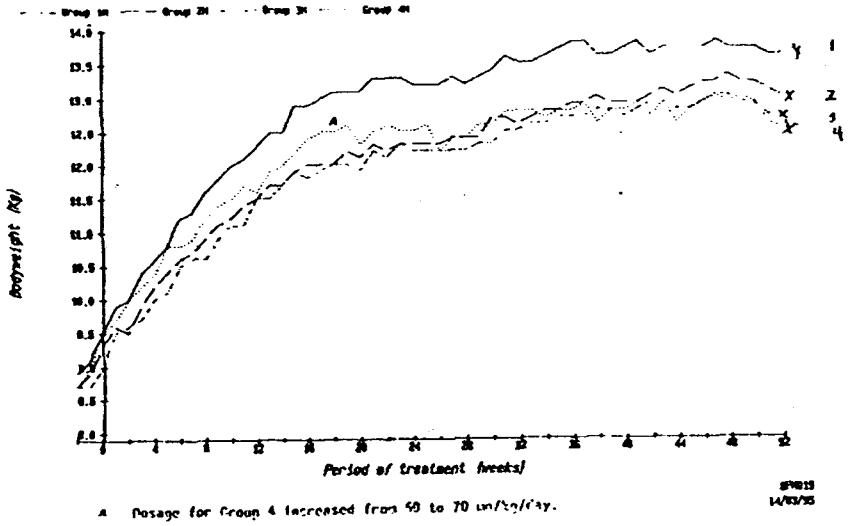


FIGURE 2A
Group mean bodyweight versus period of treatment - males

Group	:	1	2	3	4
Compound	:	Control	— SDZ HTF 919 —		
Dosage (mg/kg/day)	:	0	5	15	60/70



A Dosage for Group 4 increased from 60 to 70 mg/kg/day.

4. Food Consumption: The food consumption for control males and females were 400 and 376-400 g/animal/day, respectively. The food consumption was significantly reduced during the entire treatment period in the high dose females (6-7%) as compared to the control.

5. Hematology: There were no treatment related changes.

6. Clinical Chemistry: Alkaline phosphatase and glutamate dehydrogenase were significantly increased by 56-100% mainly in the high dose females during weeks 5, 25, 38 and 51. Plasma concentration of albumin (10-14%) and ratio of albumin to globulin (7-23%) were slightly lower in the high dose group during most of the sampling occasions. Other sporadic changes may not be considered treatment related.

7. Urinalysis: There were no treatment related changes.

8. Ophthalmoscopic Examination: There were no treatment related changes.

9. ECG: There were no treatment related changes.

10: Blood Pressure: There were no treatment related changes.

11. Organ Weights: There were no treatment related changes.

12. Gross Pathology: There were no treatment related changes.

13. Microscopic Pathology: There were no treatment related changes.

14. Toxicokinetics: The plasma concentration of the test drug was increased with the doses. The mean plasma AUCs were not markedly differed between males and females. There were no apparent difference in the plasma concentration over time. The information of plasma concentrations of the test drug were summarized in tables 7.1 and 7.2 on pages 0545 and 0546 in volume 18.5 and these tables are attached below.

SDZ HTF 919 in Beagle Dogs Following a Single Oral ——— dose of SDZ HTF 919

Sex	M[1]	F[1]	All[1]	M[2]	F[2]	All[2]	M[3]	F[3]	All[4]
Dose (mg/kg/day)	5	5	5	15	15	15	60	60	60
Time (h)	Plasma concentrations of SDZ HTF 919 (ng/ml) ; Mean ± SD								
0	-	-	-	-	-	-	-	-	-
0.5	140 ± 243	29 ± 31	84 ± 171	107 ± 117	53 ± 92	80 ± 102	260 ± 613	199 ± 255	230 ± 449
1	244 ± 357	133 ± 116	188 ± 253	394 ± 276	148 ± 182	271 ± 253	400 ± 497	501 ± 340	451 ± 409
2	240 ± 337	84 ± 57	162 ± 239	452 ± 188	265 ± 151	358 ± 187	634 ± 556	571 ± 326	602 ± 436
4	47 ± 35	21 ± 11	34 ± 27	316 ± 233	336 ± 120	326 ± 172	522 ± 570	535 ± 298	528 ± 434
7	12 ± 10	26 ± 34	19 ± 24	52 ± 11	70 ± 21	61 ± 18	392 ± 335	159 ± 87	275 ± 263
24	-	-	-	7 ± 5	5 ± 3	6 ± 4	23 ± 14	24 ± 21	24 ± 17
C_{max} (ng/ml)	266 ± 340	152 ± 91	209 ± 238	529 ± 224	392 ± 78	460 ± 172	1007 ± 440	743 ± 255	875 ± 370
t_{max} (h)	2.0 ± 1.4	2.5 ± 3.0	2.3 ± 2.2	2.0 ± 1.4	2.8 ± 1.5	2.4 ± 1.4	3.8 ± 2.8	2.3 ± 1.4	3.0 ± 2.2
AUC_{0-24} (ng.h/ml)	844 ± 1072	574 ± 196	709 ± 728	2399 ± 754	2109 ± 268	2254 ± 546	6796 ± 2840	4464 ± 1883	5630 ± 2601

\- Values below the limit of quantification ———
1 n = 4
2 n = 8
3 n = 6
4 n = 12

Table 7.1

SDZ HTF 919 in Beagle Dogs Following SDZ HTF 919 Oral ——— administration for 52 weeks

Sex	M[1]	F[1]	All[1]	M[2]	F[2]	All[2]	M[3]	F[4]	All[5]
Dose (mg/kg/day)	5	5	5	15	15	15	70	70	70
Time (h)	Plasma concentrations of SDZ HTF 919 (ng/ml) ; Mean ± SD								
0	-	-	-	7 ± 6	8 ± 2	7 ± 4	34 ± 20	66 ± 77	48 ± 54
0.5	58 ± 71	92 ± 70	75 ± 68	102 ± 83	61 ± 48	82 ± 66	55 ± 65	235 ± 244	137 ± 187
1	145 ± 97	204 ± 136	175 ± 114	305 ± 255	205 ± 195	255 ± 217	419 ± 369	476 ± 467	445 ± 395
2	177 ± 69	220 ± 92	198 ± 79	338 ± 163	210 ± 145	274 ± 158	812 ± 385	785 ± 596	799 ± 465
4	47 ± 22	47 ± 15	47 ± 17	238 ± 252	211 ± 95	225 ± 177	988 ± 664	1008 ± 713	997 ± 651
7	16 ± 6	17 ± 4	16 ± 5	73 ± 33	67 ± 32	70 ± 30	814 ± 602	931 ±810	867 ± 669
24	-	-	-	9 ± 3	6 ± 4	8 ± 4	41 ± 31	61 ± 41	50 ± 35
C_{max} (ng/ml)	199 ± 63	244 ± 115	222 ± 89	448 ± 222	317 ± 141	382 ± 196	1089 ± 991	1143 ± 747	1114 ± 831
t_{max} (h)	1.8 ± 0.5	1.5 ± 0.6	1.6 ± 0.5	2.3 ± 1.3	2.8 ± 1.5	2.5 ± 1.3	3.7 ± 0.8	4.4 ± 2.5	4.0 ± 1.7
AUC_{0-24} (ng.h/ml)	677 ± 182	814 ± 315	746 ± 249	2191 ± 672	1754 ± 204	1972 ± 516	12532 ± 8255	14010 ± 10843	13303 ± 9038

\- Values below the limit of quantification ———
1 n = 4
2 n = 8
3 n = 6
4 n = 5
5 n = 11

Table 7.2

In summary, SDZ HTF 919 was given to dogs ——— at 0, 5, 15 and 60/70 mg/kg/day for 52 weeks. The high dose of 60 mg/kg/day was increased to 70 mg/kg/day in week 18 due to lack of toxicity. Major treatment related changes were mainly in the high dose group and these included clinical signs of toxicity (salivation), decreased terminal body weight gains (13-27%) and increased activities of alkaline phosphatase and glutamate dehydrogenase (56-100%).

SPECIAL TOXICITY:

Local intravenous tolerance study in rabbits
(73LT/RB)

Testing Laboratories: Sponsor's laboratory

Study Start and Completion Dates: December 16, 1992 and May 11, 1993

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Males (2.5-3.5 kg, 3-4 months)
Females (2.5-3.5 kg, 3-4 months)
Cross-bred New Zealand White rabbits
(SPF, CRL:KBL (NZW)BR)

Methods: To determine the local irritation potential of HTF 919, HTF 919 (ampoule solution, 0.1 mg/ml or 0.033 mg/ml) was given via the partially isolated marginal ear veins of the rabbits at a rate of 0.5 ml/minute for total 4 minutes. The total amount of drug administered was 0.019-0.027 mg/kg or 0.057-0.08 mg/kg. There were 4 injection sites. Evaluations of the injection sites for inflammation, swelling, vein thrombosis and necrosis were conducted 24, 48 hours and 7 days after the injection.

Results: One rabbit in the high dose group had a minor local irritation at the injection site on the 7th day after injection. One rabbit in the low dose group displayed an inflammation and a swelling along the marginal vein at 24 hours after the injection. This study has no relevance to the proposed mode of administration (oral) in human.

Primary Skin Irritation/Corrosion study in rabbits
(Project #232796)

Testing Laboratories: [redacted]

Study Start and Completion Dates: March 17, 1998 and May 27, 1998

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Methods: To determine the skin irritation/corrosion, HTF 919 (0.5 g, moistened) was applied to clipped skin using a semi-occlusive dressing in three rabbits. Skin reaction was assessed at 1, 24, 48, and 72 hours after exposure.

Results: There was no evidence of skin irritation or corrosion observed.

Assessment of Contact Hypersensitivity in guinea pigs
(Project #232807)

Testing Laboratories: [redacted]

Study Start and Completion Dates: March 17, 1998 and July 20, 1998

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Methods: To assess the contact hypersensitivity of HTF 919, guinea pigs were intradermally injected with HTF 919 at 0.05% in corn oil on day 1. The dermal reaction was assessed on day 3. These animals were epidermally exposed to 10% sodium-dodecyl-sulfate on day 7 and to HTF 919 at 50% in corn oil on day 8. Control animals received similar treatment with corn oil only. All animals were challenged by epidermal exposure with 50% test article or vehicle two weeks after the epidermal application and skin reaction was then assessed.

Results: Skin reactions (grades 1-3) were observed in all treated animals but not in the control animals. The results suggest that HTF 919 may cause sensitization by skin contact.

CARCINOGENICITY:

A 13-Week Oral (in feed) Dose Ranging Toxicity Study in Mice
(177DFM)

Testing Laboratories: Sponsor's laboratory

Study Start and Completion Dates: February 16, 1994 and March 6, 1997

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Males (29-39 g, 8 weeks)
Females (21-28 g, 8 weeks)
SPF CD-1 Crl:CD1(ICR)BR Charles River WIGA mice

Methods: To determine the toxic and tolerable dose range of SDZ HTF 919 for the carcinogenicity study in mice, SDZ HTF 919 was given to mice (10/sex/group) in feed at 0, 150, 300, 600 and 900 mg/kg/day for males and 300, 600, 900 and 1200 mg/kg/day for females for 13 weeks. The dose selection was based on the results of a pilot 2-week dietary study in mice (176DFM). In this study, the only treatment related change was body weight loss (6-9%) in males at high dose of 800 mg/kg/day and there were no treatment related changes at doses of 200, 500 and 600 mg/kg/day in males and at doses up to 800 mg/kg/day in females. Therefore, the doses of 150, 300, 600 and 900 mg/kg/day and 300, 600, 900 and 1200 mg/kg/day were selected for the males and females, respectively. Clinical signs of toxicity and mortality were observed daily. Body weights and food consumption were determined weekly. Hematology and clinical chemistry were determined at termination. All animals were necropsied at termination and the organs (brain, liver, kidney and heart) were weighed. Gross and microscopic examinations were then conducted. The tissues examined were listed on page 11 in volume 18.2 and this list is attached below.

3.5.4 Tissue Sampling and Staining

Representative specimens from the organs listed below were collected and fixed in 10 % buffered formalin, with the exception of the testes which were fixed in Bouin's. Half of the liver lobes were deep-frozen at -18°C for possible biochemical evaluation.

Adrenals	Ovaries/oviducts
Aorta (thoracic)	Pancreas
Bones: femur/knee-joints/tibia	Pituitary
Brain	Preputial gland (male/female)
Esophagus	Prostate
Eyes with lens/optic nerve	Salivary glands
Harderian glands	Sciatic nerves
Gall bladder	Seminal vesicles
Heart with aortic arch	Skin/mammary gland
Intestine, small:	Spinal cord
duodenum, jejunum, ileum	Spleen
Intestine, large:	Sternum
cecum, colon, rectum	Stomach
Kidneys	Testes/Epididymides
Larynx	Thymus
Liver, all lobes (only 2 lobes examined)	Thyroid/Parathyroids
Lung, all lobes (only 2 lobes examined)	Tongue
Lymph nodes:	Trachea
tracheobronchial	Urinary bladder
mandibular	Uterus (body, horns, cervix)
mesenteric	Vagina
Muscles (biceps femoris)	
Nasal cavities (not examined)	All gross lesions

The toxicokinetic profile was determined in satellite animals (9/sex/group) at 1 pm, 9 pm and 5 am on treatment days 29/30 and 92/93.

Results:

1. Clinical Signs: The major clinical signs of toxicity were observed in the high dose group (males: 900 mg/kg/day; females: 1200 mg/kg/day). These included slight tremor, hunched position, pale extremities and ears, hair loss, rough coat, skin and tail necrosis, wet genital region and diarrhea. Ataxia, bluish discolored paw, ptosis and conjunctivitis were also seen in the high dose females.

2. Mortality: Three high dose males and 2 high dose females died. One female was sacrificed due to severe keratitis and panophthalmitis.

3. Body Weight: The initial and final body weights in the control group were 35 ± 2 and 37 ± 6 (males) and 24 ± 2 and 29 ± 3 (Females) g, respectively. Body weight loss was seen in males treated at 900 mg/kg/day (23%) during weeks 1 and 2 and females treated at 1200 mg/kg/day (4%) during week 1. Body weight loss (6%) was also seen during week 2 in males treated at 600 mg/kg/day. There were no treatment related changes in terminal body weight gains except the high dose males did not gain any weight up to terminal day. The body weight information is summarized in the following table.

Mean body weights (g).

	Control	Low Dose	Mid Dose 1	Mid Dose 2	High Dose
Males					
Initial	35	34	34	34	35
13 weeks	37	38	37	36	35
Females					
Initial	24	24	24	25	25
13 weeks	29	29	31	31	32

Control, low, mid 1, mid 2 and high doses = 0, 150, 300, 600 and 900 mg/kg/day for males and 0, 300, 600, 900 and 1200 mg/kg/day for females, respectively.

4. Food Consumption: The food consumption for control males and females were 4.7-5.7 and 4-5.7 g/animal/day, respectively. The food consumption was significantly reduced mainly during the first week in males treated at 600 (20.5%) and 900 (44%) mg/kg/day and females treated at 600 (8.8%), 900 (18%) and 1200 (29%) mg/kg/day as compared to the control. The reduction of food consumption was also seen during the entire study in the high dose males. The drug intake was presented in a table on page 14 in volume 18.2 and this table is attached below.

mg/kg/day	Males	Females
Controls	0	0
150	156±15	--
300	310±16	306±24
600	616±46	605±57
900	936±74	906±63
1200	--	1196±82

5. Hematology: The platelet count was slight but significantly higher in the treated males (12-21%) and high dose females (24%) than that in control animals.

6. Clinical Chemistry: Major treatment related change was a significant increase in the alanine aminotransferase (ALT) in the high dose males (186%) and females (214%). ALT was also significantly increased in the females treated at 900 mg/kg/day.

7. Organ Weights: The relative organ weights to body weight of brain (9.8%) and kidney (15%) were slightly lower in high dose females as compared to the control.

8. Gross Pathology: Following observations were found in the dead or sacrificed high dose animals: emaciation (1 male, 2 females), small spleen (3 males, 3 females), and "thymus not identified" (3 males, 2 females).

9. Microscopic Pathology: Lymphoid atrophies of thymus (3 males, 2 females) and spleen (3 males, 3 females) were found in the high dose group. The weights of thymus and spleen were not determined. Sponsor considered these changes as secondary to inanition.

10. Plasma Level of Test Drug: Mean plasma concentrations were increased with the doses. The mean plasma concentrations were summarized in a table on page 53 in volume 18.2. This table is attached below.

Mean Concentrations of HTF 919 of the Different Dose Groups Versus Day of Treatment and Gender (ng/ml)

Dose	Male			Female			Mean
mg/kg/day	day 29/30	day 92/93	Mean	day 29/31	day 92/94	Mean	Male + Female
150	23.7	20.5	22.1				
300	65.3	62.3	63.8	93.7	78.5	86.6	74.9
600	65.9	62.9	64.4	113.4	104.7	109.1	87.4
900	92.8	79.5	85.6	111.6	114.9	113.2	102
1200				176.4	81.4	128.9	

The plasma concentrations in females were higher than those in males by a factor of 1.3-1.7. The mean plasma concentrations measured on days 29/30 and 92/93 are comparable, suggesting that the drug was not accumulated over time.

In summary, in the 13-week oral dose ranging study in mice, SDZ HTF 919 was given to mice in feed at 0, 150, 300, 600 and 900 mg/kg/day for males and 300, 600, 900 and 1200 mg/kg/day for females for 13 weeks. High dose was lethal for both males and females. The major treatment related changes were clinical signs of toxicity (tremor, hunched position, pale extremities and ears, hair loss, rough coat, skin and tail necrosis, wet genital region and diarrhea), body weight loss, reduction of food consumption, increase in ALT and atrophies of thymus and spleen in the high dose group. Since high doses tested (900 mg/kg/day for males or 1200 mg/kg/day for females) were lethal and there were some toxicities seen at these dose levels, MTD can be identified between 600 and 900 mg/kg/day for males and between 900 and 1200 mg/kg/day for females. Selection of doses of 60, 200 and 600 mg/kg/day for the 2-year carcinogenicity study in mice appears adequate.

Addendum: Due to the increased incidence of adenocarcinoma in the small intestine in the mouse carcinogenicity study, sponsor re-assessed the histopathological sections of the small intestine in the following studies.

(1) The 2-week oral (in feed) dose ranging study in mice (176DFM): The results indicated that treatment with HTF 919 produced the mucosal hypertrophy and hyperplasia of the jejunal mucosa in a dose dependent manner. The slight to moderate mucosal hypertrophy and hyperplasia were identified in 2 females at 400 mg/kg/day, 4 females at 600 mg/kg/day, and 3 males and 4 females at 800 mg/kg/day (none in the control and low dose groups). There were a control and 4 treatment groups (200, 400, 600, and 800 mg/kg/day). There were 4 animals/sex/group.

(2) The 13-week oral dose ranging study in mice (177DFM): The results indicated that treatment with HTF 919 increased the incidence of the mucosal hypertrophy and hyperplasia of the small intestine in a dose dependent manner. The results are summarized in the following tables.

Incidence of hypertrophy/hyperplasia of the small intestine in males

	Control	150 mg/kg	300 mg/kg	600 mg/kg	900 mg/kg
Minimal/slight	1	1	5	3	3
Moderate	0	0	3	5	4
Total	1	1	8	8	7

Incidence of hypertrophy/hyperplasia of the small intestine in females

	Control	300 mg/kg	600 mg/kg	900 mg/kg	1200 mg/kg
Minimal/slight	0	1	5	3	4
Moderate	0	0	2	6	5
Total	0	1	7	9	9

13-Week Oral (in feed) Exploratory Toxicity Study in Mice
(971076)

Testing Laboratories: Sponsor's laboratory

Study Start and Completion Dates: February 12, 1998 and
July 14, 1999

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement. Sponsor also stated that "the study did not fully comply with GLP principles inasmuch as the determination of intestinal diamine oxidase (DAO) activity and HTF 919 in the content, mucosa and tissue of small intestine were not performed according to GLP".

Animals: Males (27-39 g, ~8 weeks)
CRL(CD1)mice

Methods: To assess the effects on the intestinal cell proliferation of SDZ HTF 919, HTF 919 was given to mice (10 males/group) in feed at 0, 200, and 600 mg/kg/day for 13 weeks. Additional ten animals per group were kept for 4 or 8 weeks after treatment for recovery. Clinical signs of toxicity and mortality were observed daily. Body weights and food consumption were determined weekly. At termination, following parameters were determined: mucosa cell kinetics of the jejunum (cell proliferation), intestinal diamine oxidase (DAO) activity, histopathology of the small intestine and liver, and plasma and tissue (small intestine) levels of HTF 919. The intestinal cell proliferation was assessed using [redacted] of bromodeoxyuridine (BrdU) incorporation. BrdU labeling index (LI) was calculated as percentage of immunoreactive nuclei/300 nuclei for each jejunum.

Results:

1. Clinical Signs: Hyperactivity was seen on the first day of treatment at 600 mg/kg/day.

2. Mortality: There were no treatment related deaths. Two control animals were sacrificed due to poor health condition. These animals had laceration and red discoloration of the ears.

3. Body Weight: The initial and final body weights in the control group were 33 ± 3 and 40 ± 4 g, respectively. Body weight loss was seen at 600 mg/kg/day during first three weeks. The terminal body weight gain was 7, 7, and 4 g in the control, 200 and 600 mg/kg/day groups, respectively.

4. Food Consumption: The food consumption was 4.7-6, 5-7, and 5-7.9 g/animal/day in the control, 200, and 600 mg/kg/day groups, respectively .

5. Mucosal Cell Kinetics: BrdU labeling index (LI) was significantly increased in the treated animals, suggesting that treatment with HTF 919 increased cell proliferation in small intestine. BrdU LI was 26.3, 30.2, and 35.2% in the control, 200, and 600 mg/kg/day groups, respectively. However, this increase was not seen in the recovery animals.

6. Intestinal Diamine Oxidase (DAO): There was significant decrease in the DAO activity in the jejunum in the treatment groups as compared to the control at the end of treatment period. This decrease was not seen at the end of recovery periods. These results were presented in a table on page 5-28.8 in volume 1.34. This table is attached below.

Table 1 Summary of the diamine oxidase activities of the main and recovery study

HTF919 Treatment	13 Week		13 Week + 4 Week Recovery		13 Week + 8 Week Recovery	
	Mean	SD	Mean	SD	Mean	SD
Group 1						
0 mg/kg/day	52.8	3.3	52.1	3.5	52.7	3.0
Group 2						
200 mg/kg/day	17.8**	5.5	51.0	4.8	51.5	3.2
Group 3						
600 mg/kg/day	15.0**	3.4	51.8	2.1	55.1	2.0

DAO activity in the table is expressed as arbitrary units/min/mg Protein.

**) Statistically significantly versus controls. $p < 0.01$.

DAO is a key enzyme in the regulation of polyamine metabolism and a known regulator of cell proliferation. Inhibition of DAO activity is expected to increase polyamine concentrations and possibly promotes cell proliferation (Agents and Actions, 23 (3/4):354-356, 1988).

7. Gross Pathology: No treatment related changes were noted.

8. Microscopic Pathology: Mucosal hyperplasia was observed at termination of 13 weeks in the jejunum at 600 mg/kg (4/10, Minimal to slight) and in the ileum at 200 mg/kg (2/10, minimal), and at 600 mg/kg (4/10, minimal to slight). The mucosal hyperplasia was characterized by elongation of the villi and increased basophilia and nuclear crowding in the intestinal crypt. The mucosal hyperplasia was not present at 4 or 8 week recovery.

9. Plasma and Tissue Level of Test Drug: Plasma level of HTF 919 was summarized in Table 4.6-1 on page 5-24 in volume 1.34 and this table is attached below.

Table 4.6-1.: Summary of toxicokinetic parameters for HTF919 in plasma

Dose	AUC_{0-24} (ng x h/mL)		AUC_{0-24}/Dose (ng x h/mL)/(mg/kg/day)	
	Days 29/30	Days 93/94	Days 29/30	Days 93/94
0 mg/kg/day	71.1	7.22		
200 mg/kg/day	462	396	2.31	1.98
600 mg/kg/day	1610	878	2.68	1.46

Measurable plasma level of HTF 919 was determined in some control animals. Sponsor stated that this may be caused by contamination during the analytic procedures. Level of HTF 919 in the jejunum and ileum ranged from ——— µg/mg mucosa. The individual level of HTF 919 highly varied.

In summary, in the 13-week oral (in feed) exploratory toxicity study in mice, HTF 919 was given to male mice in feed at 0, 200, and 600 mg/kg/day for 13 weeks. The major treatment related changes were clinical signs of toxicity (hyperactivity) and body weight loss in the high dose group. Histopathological examination revealed mucosal hyperplasia in the jejunum at 600 mg/kg (4/10, Minimal to slight) and in the ileum at 200 mg/kg (2/10, minimal), and at 600 mg/kg (4/10, minimal to slight) at week 13. The mucosal hyperplasia was not present at 4 week recovery. There were significant increase in BrdU labeling index and decrease in DAO activity in the jejunum in the treated animals in a dose dependent manner. These changes were not seen at the end of recovery period.

FDA CDER CARCINOGENICITY ASSESSMENT COMMITTEE (CAC/CAC-EC)
RODENT CARCINOGENICITY FACTSHEET

NDA: 21,200
CAS #:
DIVISION(s): HFD-180
DRUG NAME(S): SDZ HTF 919

SPONSOR: Novartis Pharmaceuticals Corporation

LABORATORY: [redacted]

P/T REVIEWER(s): Ke Zhang
P/T REVIEW DATE: June 6, 2000
CARCINOGENICITY STUDY REPORT DATE: January 23, 1998

THERAPEUTIC CATEGORY: Treatment of constipation-prone irritable bowel syndrome.

PHARMACOLOGICAL/CHEMICAL CLASSIFICATION: 5-HT_4 receptor agonist

PRIOR FDA DOSE CONCURRENCE (Div./CAC)? (Y/N; Date): No

MUTAGENIC/GENOTOXIC (Y/N/EQUIVOCAL/Na; assay): Negative in *in vitro* chromosomal aberration tests in Chinese hamster V79 cells, a forward mutation assay at HGPRT locus in Chinese hamster V79 cells, mouse bone marrow micronucleus test and unscheduled DNA synthesis (UDS) test in the rat hepatocytes. The result of Ames test was considered "equivocal" since it was positive in strain 1538 but the result could not be reproduced.

MOUSE CARCINOGENICITY STUDY (multiple studies? Std1, Std2 etc):

MOUSE STUDY DURATION (weeks): 104
STUDY STARTING DATE: September 2, 1994
STUDY ENDING DATE: January 23, 1998
MOUSE STRAIN: CD-1 mice
ROUTE: Diet
DOSING COMMENTS:

No. Mice in control1 (con1): 60
Control2 (con2): 60
Low Dose (LD): 60
Middle Dose (MD): 60
High Dose (HD): 60

Mouse Dose Levels: (mg/kg/day)

Mouse Low Dose: 60 Mouse Middle Dose: 200
Mouse High Dose: 600

Basis for doses selected (MTD, AUC ratio, saturation, maximum feasible): MTD.

MOUSE CARCINOGENICITY (negative, positive, MF, M, F): Positive (MF)

MOUSE TUMOR FINDINGS: Yes

MOUSE STUDY COMMENTS: In the 2-year dietary carcinogenicity study in mice, mice were treated with SDZ HTF 919 at 0, 60, 200 and 600 mg/kg/day for 2 years. The dose selection was based on findings from the 13 week dietary dose ranging study in mice (177DFM). High dose (900 mg/kg/day in males and 1200 mg/kg/day in females) was lethal. MTD was identified between 600 and 900 mg/kg/day for males and between 900 and 1200 mg/kg/day for females and thus selection of doses of 60, 200 and 600 mg/kg/day for the 2-year carcinogenicity study in mice appears adequate. In the current study, survival was not affected by the treatment. The terminal body weight was 96, 93 and 81.4% (males) or 94.3, 90.3 and 78% (females) of the control in the low, mid and high dose groups, respectively, suggesting that the high dose of 600 mg/kg/day exceeded MTD. The treatment with SDZ HTF 919 at high dose produced mucosal hyperplasia (8 males and 7 females) and adenocarcinoma (6 males and 2 females) in the small intestine in the high dose group (none in the control, low and mid dose groups). The high dose (600 mg/kg/day or 1800 mg/m^2/day) is ~203 times the proposed clinical dose (12 mg/day or 0.24 mg/kg/day if 50 kg body weight assumed or 8.88 mg/m^2/day). The ratio of AUC values of mouse [redacted] ng.h/ml at 600 mg/kg/day at week 4) to human (20.1 ng.h/ml at 12 mg/day) are [redacted] This study is acceptable. In conclusion, therefore, SDZ HTF 919 produced adenocarcinoma in the small intestine at high dose of 600 mg/kg/day which is ~203 times the proposed clinical dose based on body surface area.

COVERSHEET FOR CARCINOGENICITY STUDY IN MICE

1. No. Of Studies: One

2. Name of Laboratory: [redacted]

3. Strain: CD-1 mice

4. No/sex/group: 60

5. Doses (01, 02, L, M, H): 0, 0, 60, 200 and 600 mg/kg/day

6. Basis for Dose Selection Stated: Yes

7. Interim Sacrifice: No

8. Total Duration (weeks): 104

9. Week/Site for First Tumor:

Group	Male	Female
01	Week 30/lymphoma, Hematopoietic	Week 36/lymphoma, Hematopoietic
02	Week 51/histocytic sarcoma, Hematopoietic	Week 24/lymphoma, Hematopoietic
L	Week 32/lymphoma, Hematopoietic	Week 66/osteoma, femur/marrow
M	Week 62/histiocytic sarcoma, Hematopoietic	Week 45/lymphoma, Hematopoietic
H	Week 44/lymphoma, Hematopoietic	Week 50/histiocytic sarcoma, Hematopoietic

01 = control1, 02=control2, L, M and H = 60, 200 and 600 mg/kg/day, respectively

10. No. Alive at Termination:

Sex	Males					Females				
Group	con1	con2	low	mid	high	con1	con2	low	mid	high
No. alive	31	39	27	35	38	25	30	24	31	34
% survival	52	65	45	58	63	42	50	40	52	57

0 = control1, 01 = control2, L=60 mg/kg/day, M = 200 mg/kg/day, H = 600 mg/kg/day

11. Statistical Methods used: The tumor data were analyzed using the prevalence method of Peto (Peto, R. et.al., Guidelines for simple, sensitive significance tests for carcinogenic effects in long-term animal experiment in Long-term and short term screening assays for carcinogens: a critical appraisal. Geneva: WHO, pp 311-426, 1980).

12. Attach Tumor and Non-tumor Data for Each Tissue: Tumor and non-tumor data attached in Appendix I.

Two Year Carcinogenicity Study of HTF 919 in Diet in Mice
(— 034/970331)

Testing Laboratories: [redacted]

Study Start and Completion Dates: August 16, 1994 and
January 23, 1998

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Males (25-35 g, 5-6 weeks)
Females (20-31 g, ~7 weeks)
CD-1 mice from [redacted]

Methods: To determine the carcinogenic potential of SDZ HTF 919, mice (60/sex/group) were treated with SDZ HTF 919 in diet at 0, 60, 200 and 600 mg/kg/day for 2 years. The study design was summarized in a table on page 0015 of volume 18.6 and this table is attached below.

Group	Treatment	Dosage (mg/kg/day)	Animal numbers: Main study Male	Main study Female	Satellite study† Male	Satellite study† Female
1	Control	0	60	60	16	16
2	Control	0	60	60	-	-
3	SDZ HTF 919	60	60	60	16	16
4	SDZ HTF 919	200	60	60	16	16
5	SDZ HTF 919	600	60	60	16	16

The dose selection was based on findings from the 13 week dietary dose ranging study in mice (177DFM). MTD was identified between 600 and 900 mg/kg/day for males and 900 and 1200 mg/kg/day for females and selection of doses of 60, 200 and 600 mg/kg/day for the 2-year carcinogenicity study in mice appears adequate. In this 2-year carcinogenicity study, clinical signs of toxicity and mortality were observed daily. Body weights and food consumption were determined weekly. Ophthalmoscopy was performed before the treatment and at termination. Hematology was conducted at termination. All animals were necropsied at termination. Gross and histopathological examination were performed. The tissues examined were listed on pages 0023 and 0024 in volume 6 and the list is attached below.

Adrenals	Oesophagus
Aorta	Ovaries with oviduct
Brain	Pancreas
Caecum	Pituitary
Colon	Prostate
Duodenum	Rectum
Epididymides	Salivary gland
Eyes and optic nerves	- submandibular, left
Femoral bone and marrow	Sciatic nerve, left
Gall bladder	Seminal vesicles
Harderian glands	Skeletal muscle - thigh
Heart	Skin
Ileum	Spinal cord
Jejunum	Spleen
Kidneys	Sternum and marrow
Larynx	Stomach - keratinised
Liver	- glandular
Lungs with mainstem bronchi	Testes
Lymph nodes - mandibular	Thymus
- mesenteric	Thyroid with parathyroids
- bronchial	Trachea
Mammary gland - caudal	Urinary bladder
Nasal turbinates	Uterus with cervix
	Vagina.

Plasma levels of the test drug were determined in weeks 4, 48, 70, 90 and 104 in the satellite animals (16/sex/group). The blood samples were collected for determination of plasma levels of test drug at midnight, 8:00 am and 4:00 pm. The tumor data were analyzed using the prevalence method of Peto (Peto, R. et.al., Guidelines for simple, sensitive significance tests for carcinogenic effects in long-term animal experiment in Long-term and short term screening assays for carcinogens: a critical appraisal. Geneva: WHO, pp 311-426, 1980).

Results:

1. Clinical Signs: The major treatment related change was piloerection and ungroomed coat mainly in the high dose group. The incidence of piloerection was 29-31, 34, 33 and 40 (males) or 26, 30, 33 and 33 (females) in the control, low, mid and high dose groups, respectively. The incidence of ungroomed coat was 7-10 and 16 (males) or 8-16 and 23 (females) in the control and high dose groups, respectively.

2. Mortality: The mortality rate was not affected by the treatment. The intercurrent mortality (unscheduled deaths) was summarized in the following table.

Mortality (unscheduled deaths)

	Males					Females				
Weeks	Con1	Con2	Low	Mid	High	Con1	Con2	Low	Mid	High
0-52	4	3	2	4	2	3	5	2	3	2
53-77	2	1	9	4	2	8	6	11	6	6
78-90	8	6	5	11	7	8	7	10	8	7
91-104	12	9	14	4	10	14	11	13	12	11
Total	26	19	30	23	21	33	29	36	29	26

Con1=control1, con2=control2, low, mid and high = 60, 200 and 600 mg/kg/day, respectively

3. Body Weight: The initial and final body weights for the control animals were 31 and 47 g for males or 26 and 43.7 g for females. The terminal body weight was 96, 93 and 81.4% (males) or 94.3, 90.3 and 78% (females) of the control in the low, mid and high dose groups, respectively, suggesting that the high dose of 600 mg/kg/day exceeded MTD. The body weight information is summarized in the following table.

Mean body weights (g)

	Control 1	Control 2	Low Dose	Mid Dose	High Dose
Males					
Initial	30.9	31.3	31.2	30.9	31.1
13 weeks	39.8	39.8	40.9	39.0	35.6
26 weeks	44.5	43.3	44.6	41.6	36.7
52 weeks	49.3	49.0	49.8	46.6	40.0
78 weeks	49.7	49.6	50.8	47.2	40.6
Terminal	47.4	47.0	45.3	43.9	38.4
Females					
Initial	26.0	25.9	25.8	26.0	25.8
13 weeks	32.3	32.0	32.0	31.1	29.4
26 weeks	34.4	34.2	34.2	33.2	30.5
52 weeks	39.3	38.2	37.1	37.2	33.3
78 weeks	40.6	40.6	38.2	37.6	34.3
Terminal	43.7	41.0	40.0	38.3	32.9

The growth curves depicted in figures 3A and 3B on pages 0046 and 0047 in volume 18.6 are attached below.

FIGURE 3A
Group mean bodyweight versus period of treatment - males

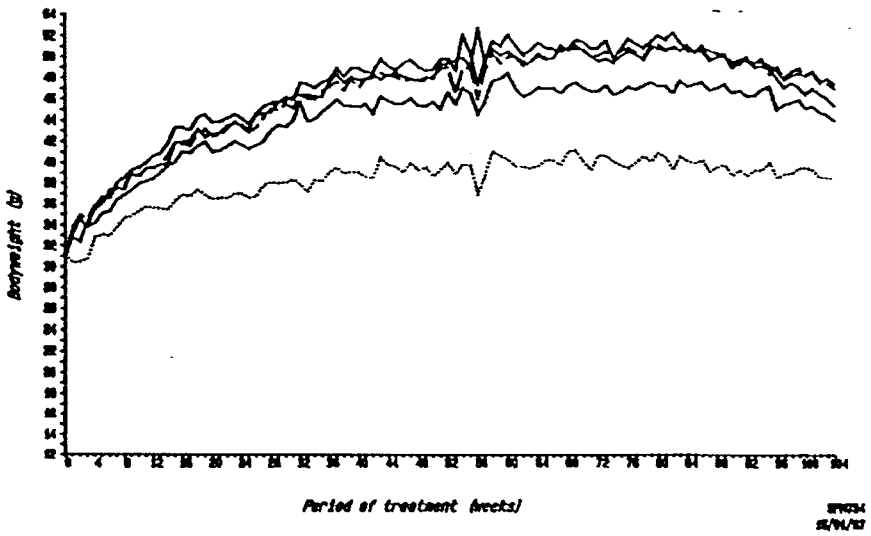
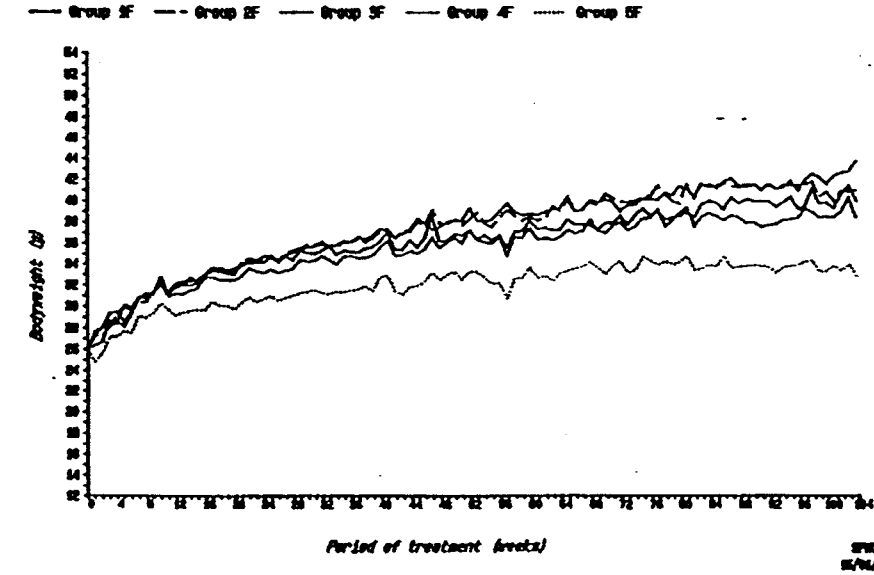


FIGURE 3B
Group mean bodyweight versus period of treatment - females



4. Food Consumption: There were no treatment related changes. Average food consumption in the control group was 4.9-6.6 (males) or 4.9-6 (females) g/mouse/day. The mean achieved doses were 60.7, 202.2 and 611.3 (males) or 60.6, 201.2 and 605.4 mg/kg/day for the low, mid and high dose groups, respectively.

5. Ophthalmoscopy: There were no treatment related changes.

6. Hematology: There were no treatment related changes.

7. Gross Pathology: Mass was found in the jejunum in 4 high dose males and 1 female. Mass was also found in ileum in 1 high dose female. These were identified as adenocarcinoma by histopathological examination.

8. Histopathology:

Non-Neoplastic Changes: Mucosal hyperplasia in the small intestine was found in the high dose group (8/60 males and 7/60 females). There was none in the control, low and mid dose groups. This was characterized by increased cellularity, elevated mitotic rate, increased basophilia and nuclear crowding in the intestine crypts.

Neoplastic Changes: The treatment with SDZ HTF 919 at high dose produced adenocarcinomas in the small intestine (6 males and 2 females, none in control, $p < 0.001$). There was no background incidence of this tumor in the historical control data from studies in CD-1 mice conducted at the testing laboratory [redacted] during 1992-1995 and from [redacted] (Spontaneous neoplastic lesions in the Crl:CD-1 (ICR)BR mouse, [redacted] 1987). One high dose male had adenocarcinoma in the colon. There was evidence of statistically significant increase in the following tumors in the low dose group: benign hepatocellular adenoma (male), pulmonary carcinoma (male), uterus benign leiomyoma and uterine polyp. However, there was no evidence for a trend and the incidences of these tumors in the mid and high dose groups were not significantly increased. Therefore, these are not considered as treatment related. This information was summarized in tables 16, 18, 19, 21, 28 and 29 on pages 243, 245, 246, 248, 255, and 256 in volume 1.21.

TABLE 16

Jejunum
Results of time-to-tumour analysis for malignant adenocarcinoma in males

Combined control groups

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison *p*-value^	Trend test *p*- value#
			Observed (O)	Expected (E)			
1+2	0	120	0	2.46	0.00		
3	60	60	0	1.09	0.00	0.500	
4	200	60	0	1.19	0.00	0.500	
5	600	60	6	1.27	4.74	0.001	<0.001

Control group 1

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison *p*-value^	Trend test *p*- value#
			Observed (O)	Expected (E)			
1	0	60	0	1.47	0.00		
3	60	60	0	1.39	0.00	0.500	
4	200	60	0	1.52	0.00	0.500	
5	600	60	6	1.62	3.70	0.025	<0.001

Control group 2

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison *p*-value^	Trend test *p*- value#
			Observed (O)	Expected (E)			
2	0	60	0	1.62	0.00		
3	60	60	0	1.35	0.00	0.500	
4	200	60	0	1.46	0.00	0.500	
5	600	60	6	1.56	3.84	0.017	<0.001

^ One-tailed pairwise comparisons against the control group(s).
\# One-tailed trend tests using control group(s) up to the respective group.

TABLE 18

Liver
Results of time-to-tumour analysis for benign hepatocellular adenoma in males

Combined control groups

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison *p*-value^	Trend test *p*- value#
			Observed (O)	Expected (E)			
1+2	0	120	24	24.96	0.96		
3	60	60	20	11.01	1.82	0.012	
4	200	60	13	12.29	1.06	0.472	
5	600	60	4	12.74	0.31	0.988	0.999

Control group 1

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison *p*-value^	Trend test *p*- value#
			Observed (O)	Expected (E)			
1	0	60	12	12.31	0.97		
3	60	60	20	11.37	1.76	0.043	
4	200	60	13	12.47	1.04	0.500	
5	600	60	4	12.86	0.31	0.982	0.999

Control group 2

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison *p*-value^	Trend test *p*- value#
			Observed (O)	Expected (E)			
2	0	60	12	12.91	0.93		
3	60	60	20	11.12	1.80	0.033	
4	200	60	13	12.26	1.06	0.426	
5	600	60	4	12.71	0.31	0.969	0.999

^ One-tailed pairwise comparisons against the control group(s).
\# One-tailed trend tests using control group(s) up to the respective group.

TABLE 19

Lungs
Results of time-to-tumour analysis for malignant pulmonary carcinoma in males

Combined control groups[a]

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison p-value^	Trend test p- value#
			Observed (O)	Expected (E)			
1+2	0	120	11	11.91	0.92		
3	60	60	12	4.88	2.46	0.011	
4	200	60	4	5.94	0.67	0.606	
5	600	60	2	6.27	0.32	0.887	0.979

Control group 1

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison p-value^	Trend test p- value#
			Observed (O)	Expected (E)			
1	0	60	5	5.71	0.88		
3	60	60	12	4.99	2.40	0.037	
4	200	60	4	5.98	0.67	0.536	
5	600	60	2	6.32	0.32	0.809	0.988

Control group 2

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison p-value^	Trend test p- value#
			Observed (O)	Expected (E)			
2	0	60	6	6.44	0.93		
3	60	60	12	5.11	2.35	0.042	
4	200	60	4	6.13	0.65	0.586	
5	600	60	2	6.32	0.32	0.855	0.988

a Test for non-linearity: $p=0.006$
^ One-tailed pairwise comparisons against the control group(s).
# One-tailed trend tests using control group(s) up to the respective group.

TABLE 21

Jejunum and ileum combined
Results of time-to-tumour analysis for malignant adenocarcinoma in females

Combined control groups

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison *p*-value^A	Trend test *p*- value#
			Observed (O)	Expected (E)			
1+2	0	120	0	0.78	0.00		
3	60	60	0	0.36	0.00	0.500	
4	200	60	0	0.41	0.00	0.500	
5	600	60	2	0.44	4.53	0.125	0.007

Control group 1

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison *p*-value^A	Trend test *p*- value#
			Observed (O)	Expected (E)			
1	0	60	0	0.47	0.00		
3	60	60	0	0.45	0.00	0.500	
4	200	60	0	0.52	0.00	0.500	
5	600	60	2	0.55	3.61	0.273	0.0.18

Control group 2

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate (O/E)	Pairwise comparison *p*-value^A	Trend test *p*- value#
			Observed (O)	Expected (E)			
2	0	60	0	0.50	0.00		
3	60	60	0	0.44	0.00	0.500	
4	200	60	0	0.51	0.00	0.500	
5	600	60	2	0.54	3.67	0.258	0.0.17

^A One-tailed pairwise comparisons against the control group(s).
\# One-tailed trend tests using control group(s) up to the respective group.

TABLE 28

Uterus
Results of time-to-tumour analysis for benign leiomyoma in females

Combined control groups

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate	Pairwise comparison	Trend test
			Observed (O)	Expected (E)	(O/E)	*p*-value^A	*p*- value#
1+2	0	120	2	3.53	0.57		
3	60	60	4	1.53	2.62	0.068	
4	200	60	0	1.92	0.00	0.615	
5	600	60	3	2.02	1.48	0.268	0.322

Control group 1

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate	Pairwise comparison	Trend test
			Observed (O)	Expected (E)	(O/E)	*p*-value^A	*p*- value#
1	0	60	2	2.08	0.96		
3	60	60	4	1.93	2.07	0.311	
4	200	60	0	2.43	0.00	0.792	
5	600	60	3	2.56	1.17	0.500	0.538

Control group 2

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate	Pairwise comparison	Trend test
			Observed (O)	Expected (E)	(O/E)	*p*-value^A	*p*- value#
2	0	60	0	1.79	0.00		
3	60	60	4	1.46	2.73	0.041	
4	200	60	0	1.84	0.00	0.500	
5	600	60	3	1.91	1.57	0.135	0.257

^A One-tailed pairwise comparisons against the control group(s).
\# One-tailed trend tests using control group(s) up to the respective group.

TABLE 29

Uterus
Results of time-to-tumour analysis for benign uterine polyp in females

Combined control groups

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate	Pairwise comparison	Trend test
			Observed (O)	Expected (E)	(O/E)	*p*-value▲	*p*- value#
1+2	0	120	8	8.28	0.97		
3	60	60	10	4.13	2.42	0.030	
4	200	60	3	4.25	0.71	0.553	
5	600	60	0	4.34	0.00	0.953	0.993

Control group 1

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate	Pairwise comparison	Trend test
			Observed (O)	Expected (E)	(O/E)	*p*-value▲	*p*- value#
1	0	60	3	3.99	0.75		
3	60	60	10	3.70	2.70	0.026	
4	200	60	3	4.04	0.74	0.500	
5	600	60	0	4.27	0.00	0.900	0.995

Control group 2

Group	Dose level (mg/kg/day)	Initial group size	Number of animals with tumours		Relative tumour rate	Pairwise comparison	Trend test
			Observed (O)	Expected (E)	(O/E)	*p*-value▲	*p*- value#
2	0	60	5	4.38	1.14		
3	60	60	10	4.47	2.24	0.143	
4	200	60	3	4.54	0.66	0.663	
5	600	60	0	4.61	0.00	0.970	0.998

▲ One-tailed pairwise comparisons against the control group(s).
\# One-tailed trend tests using control group(s) up to the respective group.

The incidence of neoplastic and non-neoplastic histopathological findings were summarized in sponsor's tables 10C (pages 0155-0159) and 10F (pages 0183-0198) in volume 18.6 and these tables are attached in Appendix I.

9. Toxicokinetics: The plasma concentrations were not markedly different between males and females and there was no apparent accumulation of the test drug over time. These results were

summarized in tables 1, 2, 3, 4 and 5 on pages 271-275 in volume 1.21. These tables are attached below.

Table 1 : Plasma concentrations of SDZ HTF 919 in mice during a lifespan oral (diet) oncogenicity study - Week 4

Gender	M	F	M	F	M	F
Dose mg/kg/day	60	60	200	200	600	600
Time (h)	Plasma concentrations (ng/ml) ; mean ± SD					
0	3.95 ± 1.22	3.95 ± 1.54	35.95 ± 16.25	23.57 ± 12.33	101.68 ± 26.37	126.03 ± 16.94
8	3.50 ± 1.21	3.89 ± 2.05	36.09 ± 14.83	26.92 ± 23.47	84.15 ± 29.62	129.09 ± 32.81
16	2.95 ± 2.67	0.27 ± 0.55	15.66 ± 2.90	9.73 ± 5.18	21.62 ± 4.20	20.16 ± 10.68
AUC0-24h (ng.h/ml)	63.2	63.3	701.6	481.8	1659.8	2218.2
AUC0-24h/Dose (ng.h.ml)/(mg/kg)	1.39	1.05	3.51	2.41	2.77	3.70

n = 4

Re : Tables A2, A3, A4

Table 2 : Plasma concentrations of SDZ HTF 919 in CD-1 mice during a lifespan oral (diet) oncogenicity study - Week 48

Gender	M	F	M	F	M	F
Dose mg/kg/day	60	60	200	200	600	600
Time (h)	Plasma concentrations (ng/ml) ; mean ± SD					
0	3.86 ± 1.20	2.21 ± 0.11	30.94 ± 21.26	33.52 ± 14.52	70.67 ± 46.02	78.85 ± 49.75
8	3.01 ± 0.66	2.62 ± 0.77	12.34 ± 7.19	14.80 ± 9.01	30.50 ± 9.59	52.14 ± 27.26
16	2.19 ± 1.38	1.80 ± 0.75	9.31 ± 2.74	9.37 ± 0.97	32.50 ± 2.07	21.07 ± 6.82
AUC0-24h (ng.h/ml)	72.5	53.0	420.7	461.5	1069.4	1216.5
AUC0-24h/Dose	1.21	0.88	2.10	2.31	1.78	2.03

n = 4

Re : Tables A6, A7, A8

Table 3 : Plasma concentrations of SDZ HTF 919 in CD-1 mice during a lifespan oral (diet) oncogenicity study - Week 70

Gender	M	F	M	F	M	F
Dose mg/kg/day	60	60	200	200	600	600
Time (h)	Plasma concentrations (ng/ml) ; mean ± SD					
0	3.50 ± 0.58	3.45 ± 0.34	26.78 ± 15.58	56.36 ± 34.30	55.96 ± 28.41	58.61 ± 13.77
8	3.64 ± 1.26	4.20 ± 2.46	22.34 ± 8.77	16.76 ± 9.24	45.46 ± 10.67	36.70 ± 15.96
16	2.90 ± 1.25	2.08 ± 0.31	12.90 ± 8.57	18.93 ± 10.98	28.05 ± 3.15	31.89 ± 7.75
AUC0-24h (ng.h/ml)	81.9	77.8	496.2	752.6	1020.1	1001.6
AUC0-24h/Dose	1.37	1.30	2.48	3.76	1.70	1.67

n = 4

Re : Tables A10, A11, A12

Table 4 : Plasma concentrations of SDZ HTF 919 in CD-1 mice during a lifespan oral (diet) oncogenicity study - Week 90

Gender	M	F	M	F	M	F
Dose mg/kg/day	60	60	200	200	600	600
Time (h)	Plasma concentrations (ng/ml) ; mean ± SD					
0	4.58* ± 2.17	2.72* ± 0.82	46.91* ± 16.26	58.22* ± 22.31	49.66* ± 18.14	59.65* ± 13.95
8	1.94* ± 0.92	3.98* ± 1.56	12.61* ± 11.94	20.83* ± 18.12	30.69* ± 10.56	33.73* ± 14.92
16	4.14* ± 1.12	1.30* ± 0.15	38.10* ± 27.97	12.39* ± 12.87	28.56* ± 4.51	22.95* ± 28.45
AUC0-24h (ng.h/ml)	85.3	64.0	780.2	731.5	872.9	930.6
AUC0-24h/Dose	1.42	1.07	3.90	3.66	1.45	1.55

* n = 4
* n = 2
* n = 3

Re : Tables A14, A15, A16

Table 5 : Plasma concentrations of SDZ HTF 919 in CD-1 mice during a lifespan oral (diet) oncogenicity study - Week 104

Gender	M	F	M	F	M	F
Dose mg/kg/day	60	60	200	200	600	600
Time (h)	Plasma concentrations (ng/ml) ; mean ± SD					
0	4.29* ± 1.90	3.95* ± 3.67	22.08* ± 5.66	62.08* ± 41.79	62.40* ± 20.87	93.58* ± 40.91
8	1.61° ± 0.96	2.56* ± 0.93	12.12* ± 6.72	16.34* ± 10.55	23.81° ± 12.78	59.33° ± 30.35
16	1.85* ± 1.08	0.37* ± 0.74	9.87* ± 7.55	25.80* ± 29.59	21.86* ± 17.26	63.52* ± 54.14
AUC0-24h (ng.h/ml)	64.6	55.0	352.4	832.2	864.6	1731.4
AUC0-24h/Dose	1.08	0.92	1.76	4.16	1.44	2.89

* n = 4
* n = 5
* n = 6
* n = 7
* n = 8

Re : Tables A18, A19, A20

The ratio of AUC values of mouse [redacted] ng.h/ml at 200 mg/kg/day at week 4) to human (20.1 ng.h/ml at 12 mg/day) are [redacted] The ratio of AUC values of mouse [redacted] ng.h/ml at 600 mg/kg/day at week 4) to human (20.1 ng.h/ml at 12 mg/day) are [redacted]

In summary, in the 2-year dietary carcinogenicity study in mice, mice were treated with SDZ HTF 919 at 0, 60, 200 and 600 mg/kg/day for 2 years. The dose selection was based on findings from the 13 week dietary dose ranging study in mice (177DFM). High dose (900 mg/kg/day in males and 1200 mg/kg/day in females) was lethal. MTD was identified between 600 and 900 mg/kg/day for males and between 900 and 1200 mg/kg/day for females and selection of doses of 60, 200 and 600 mg/kg/day for the 2-year carcinogenicity study in mice appears adequate. In the current study, survival was not affected by the treatment. The terminal body weight was 96, 93 and 81.4% (males) or 94.3, 90.3 and 78% (females) of the control in the low, mid and high dose groups, respectively, suggesting that the high dose of 600 mg/kg/day exceeded MTD. The major treatment related non-neoplastic change was the mucosal hyperplasia in the small intestine in the high dose males (8/60) and females (7/60) (none in the control, low and mid dose groups). The treatment with SDZ HTF 919 produced adenocarcinoma in the small intestine in the high dose group (6 males and 2 females) (none in the control, low and mid dose groups). There was no background incidence of this tumor in the historical control data from studies in CD-1 mice conducted at the testing laboratory [redacted] during 1992-1995 and from [redacted] (Spontaneous neoplastic lesions in the Crl:CD-1 (ICR)BR mouse, [redacted] 1987). The high doses (600 mg/kg/day or 1800 mg/m^2/day) are ~203 times the proposed clinical dose (12 mg/day or 0.24 mg/kg/day if 50 kg body weight assumed or 8.88 mg/m^2/day). The ratio of AUC values of mouse ([redacted] ng.h/ml at 600 mg/kg/day at week 4) to human (20.1 ng.h/ml at 12 mg/day) are [redacted] This study is acceptable. In conclusion, therefore, SDZ HTF 919 produced adenocarcinoma in the small intestine at high dose of 600 mg/kg/day which is ~203 times the proposed clinical dose based on body surface area.

FDA CDER CARCINOGENICITY ASSESSMENT COMMITTEE (CAC/CAC-EC)
RODENT CARCINOGENICITY FACTSHEET

NDA: 21,200
CAS #:
DIVISION(s): HFD 180
DRUG NAME(S): SDZ HTF 919
SPONSOR: Norvartis
LABORATORY: [redacted]

P/T REVIEWER(s): Ke Zhang
P/T REVIEW DATE: June 6, 2000

CARCINOGENICITY STUDY REPORT DATE: March 9, 1998

THERAPEUTIC CATEGORY: Treatment of constipation-prone irritable bowel syndrome.

PHARMACOLOGICAL/CHEMICAL CLASSIFICATION: 5-HT_4 receptor agonist

PRIOR FDA DOSE CONCURRENCE (Div./CAC)? (Y/N; Date): No

MUTAGENIC/GENOTOXIC (Y/N/EQUIVOCAL/Na; assay): Negative in *in vitro* chromosomal aberration tests in Chinese hamster V79 cells, a forward mutation assay at HGPRT locus in Chinese hamster V79 cells, mouse bone marrow micronucleus test and unscheduled DNA synthesis (UDS) test in the rat hepatocytes. The result of Ames test was considered "equivocal" since it was positive in strain 1538 but the result could not be reproduced.

RAT CARCINOGENICITY STUDY (multiple studies? Std1, Std2 etc):

RAT STUDY DURATION (weeks): 110 (females) or 124 (males)
STUDY STARTING DATE: May 27, 1994
STUDY ENDING DATE: March 9, 1998
RAT STRAIN: HanIbm Wistar rats
ROUTE: Diet

DOSING COMMENTS:

No. RAT in control1 (con1): 50
Control2 (con2): 50
Low Dose (LD): 50
Middle Dose (MD): 50
High Dose (HD): 50

RAT Dose Levels (mg/kg/day)

RAT Low Dose: 20 RAT Middle Dose: 80
RAT High Dose: 180

Basis for doses selected (MTD, AUC ratio, saturation, maximum feasible): MTD

RAT CARCINOGENICITY (negative, positive, MF, M, F): Negative (MF)

RAT TUMOR FINDINGS: No

RAT STUDY COMMENTS: In the 2-year dietary carcinogenicity study in rats, SDZ HTF 919 was given to rats in diet at 0, 20, 80 and 180 mg/kg/day for 110 weeks (females) or 124 weeks (males). The dose selection was adequate based on findings in the 26-week dietary toxicity study in rats (395R). In this study, high dose of 240 mg/kg/day was above MTD since the terminal body weight gain was decreased by 22.4-36% at this dose and the dose of 180 mg/kg/day was then selected as the high dose in the carcinogenicity study. In the current study, the terminal body weight was 96.5, 88.6 and 75.5% (males) or 93, 86 and 72% (females) of the control in the low, mid and high dose groups, respectively, suggesting that the high dose of 180 mg/kg/day exceeded MTD. The food consumption was slightly lower (11-12%) in the high dose group as compared the control. The mucosal hyperplasia in the small intestine was found in 2 control males and 5 high dose animals (4 males and 1 female). In the original report, the incidence of ovarian cysts (bursal, follicular and luteal) was significantly increased in the treated females as compared to the concurrent control. Subsequent evaluation did not reveal any treatment related increase in the incidence of ovarian cysts. There were no clearly treatment related increases in the tumor incidences. The study is acceptable.

COVERSHEET FOR CARCINOGENICITY STUDY IN RATS

1. No. Of Studies: One

2. Name of Laboratory: [redacted]

3. Strain: HanIbm Wistar rats

4. No/sex/group: 50

5. Doses (0, L, M, H): 0, 20, 80 and 180 mg/kg/day

6. Basis for Dose Selection Stated: Yes

7. Interim Sacrifice: No

8. Total Duration (weeks): 110 (females) and 124 (males)

9. Week/site for First Tumor:

Group	Male	Female
0	Week 82/adenoma, pituitary	Week 75/adenoma, pituitary
0	Week 39/fibrosarcoma, skeletal muscle	Week 61/adenoma, pituitary
L	Week 54/fibrosarcoma, skin	Week 65/fibroadenoma, mammary
M	Week 50/lymphoma, Hematopioetic	Week 63/adenoma, pituitary
H	Week 73/adenoma, pituitary	Week 76/adenocarcinoma, uterus

01 = control1, 02=control2, L, M and H = 20, 80 and 180 mg/kg/day, respectively

10. No. Alive at Termination:

	Male					Female				
mg/kg/day	con	con	low	mid	high	con	con	low	mid	high
No. alive	29	27	24	27	37	25	28	25	30	37
% survival	58	54	48	54	74	50	56	50	60	74

01 = control1, 02=control2, L, M and H = 20, 80 and 180 mg/kg/day, respectively

11. Statistical Methods Used: The tumor data were analyzed using the prevalence method of Peto (Peto, R. et.al., Guidelines for simple, sensitive significance tests for carcinogenic effects in long-term animal experiment in Long-term and short term screening assays for carcinogens: a critical appraisal. Geneva: WHO, pp 311-426, 1980).

12. Attach Tumor and Non-tumor Data For Each Tissue: Tumor and non-tumor data attached in Appendix II.

Two Year Carcinogenicity Study of SDZ HTF 919 in Diet in Rats
(- 029/970357)

Testing Laboratories: [redacted]

Study Start and Completion Dates: May 27, 1994 and March 9, 1998

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Males (~231 g, 35-42 weeks)
Females (172-174 g, 35-42 weeks)
HanIbm Wistar rats

Methods: To determine the carcinogenic potential of SDZ HTF 919, rats (50/sex/group) were treated with SDZ HTF 919 in diet at 0, 20, 80 and 180 mg/kg/day. The intended duration of treatment was 104 weeks. The duration, however, was extended to 110 weeks for females and 124 weeks for males due to high survival rate. The animals in each group were sacrificed when the number of survivors reached 25. The dose selection was based on findings from the 26-week dietary toxicity study in rats (395R). The high dose (240 mg/ kg/day) was above MTD since the terminal body weight gain was decreased by 22.4-36% at this dose. Therefore, sponsor selected 180 mg/kg/day as the high dose for the carcinogenicity study. In the carcinogenicity study, clinical signs of toxicity and mortality were observed daily. Body weights and food consumption were determined weekly. Ophthalmology examination was conducted before and after 52 weeks. Hematology was performed during weeks 111 (females) and 124 (males). All animals were necropsied at termination and gross and histopathological examinations were conducted. The tissues examined were listed on page 0023 in volume 12 and this list is attached below.

Adrenals	Ovaries with oviducts
Aorta	Pancreas
Brain	Pituitary
Caecum	Prostate
Colon	Rectum
Duodenum	Salivary gland - submandibular
Epididymides	Sciatic nerves
Eye and optic nerve, one only	Seminal vesicles
Femoral bone and marrow	Skeletal muscle - thigh
Harderian glands	Skin
Head (nasal turbinates)	Spinal cord
Heart	Spleen
Ileum	Sternum
Jejunum	Stomach
Kidneys	Testes
Larynx	Thymus
Liver	Thyroid with parathyroids
Lungs with mainstem bronchi	Trachea
Lymph nodes - mandibular	Urinary bladder
- mesenteric	Uterus with cervix
Mammary gland - caudal	Vagina
Oesophagus	

Plasma levels of the test drug were determined during weeks 4, 48, 72 and 104 in the satellite animals (5/sex/group). The blood samples were collected for determination of plasma level of test drug at 6:00 am, noon, 6:00 pm and midnight. The tumor data were analyzed using the prevalence method of Peto (Peto, R. et.al., Guidelines for simple, sensitive significance tests for carcinogenic effects in long-term animal experiment in Long-term and short term screening assays for carcinogens: a critical appraisal. Geneva: WHO, pp 311-426, 1980).

Results:

1. Clinical Signs: Ungroomed coats and thin build were seen in the high dose animals.

2. Mortality: Mortality was significantly lower in the high dose group as compared to combined control. The mortality was 21-23, 26, 23 and 13 (males) or 22-25, 25, 20 and 13 (females) in the control, low, mid and high dose groups, respectively. The intercurrent mortality (unscheduled deaths) was summarized in the following table.

Mortality (unscheduled deaths)										
	Males					Females				
Weeks	Con1	Con2	Low	Mid	High	Con1	Con2	Low	Mid	High
0-54	1	1	1	1	0	0	0	0	0	0
55-77	0	3	2	2	1	2	4	2	3	1
78-90	5	3	5	3	1	3	4	4	3	2
91-104	4	2	5	2	2	14	10	10	10	6
Total	10	9	13	8	4	19	18	16	16	9

Con1=control1, con2=control2, low, mid and high = 20, 80 and 180 mg/kg/day, respectively

3. Body Weight: The initial and final body weights in the control groups were 231 and 512 g for males or 173 and 374 g for females. The terminal body weight (1-104 week) was 96.5, 88.6 and 75.5% (males) or 93, 86 and 72% (females) of the control in the low, mid and high dose groups, respectively, suggesting that the high dose of 180 mg/kg/day exceeded MTD. The body weight information is summarized in the following table.

Mean body weights (g)

	Control 1	Control 2	Low Dose	Mid Dose	High Dose
Males					
Initial	231	231	231	231	231
13 weeks	394	389	390	386	342
26 weeks	447	437	439	425	370
52 weeks	510	497	499	477	406
78 weeks	550	538	530	504	414
104 weeks	550	542	527	484	412
Terminal	512	476	497	461	405
Females					
Initial	172	174	170	171	171
13 weeks	242	245	242	240	221
26 weeks	266	270	266	258	232
52 weeks	310	318	307	281	246
78 weeks	364	376	350	313	260
104 weeks	386	380	357	330	276
Terminal	374	366	349	327	278

The growth curves were depicted in figures 3A and 3B on pages 0046 and 0047 in volume 18.12. These figures are attached below.

FIGURE 3A
Group mean bodyweight versus period of treatment - males

Group	:	1	2	3	4	5
Compound	:	Control		— SDZ HTF 919 —		
Dosage (mg/kg/day)	:	0	0	20	60	180

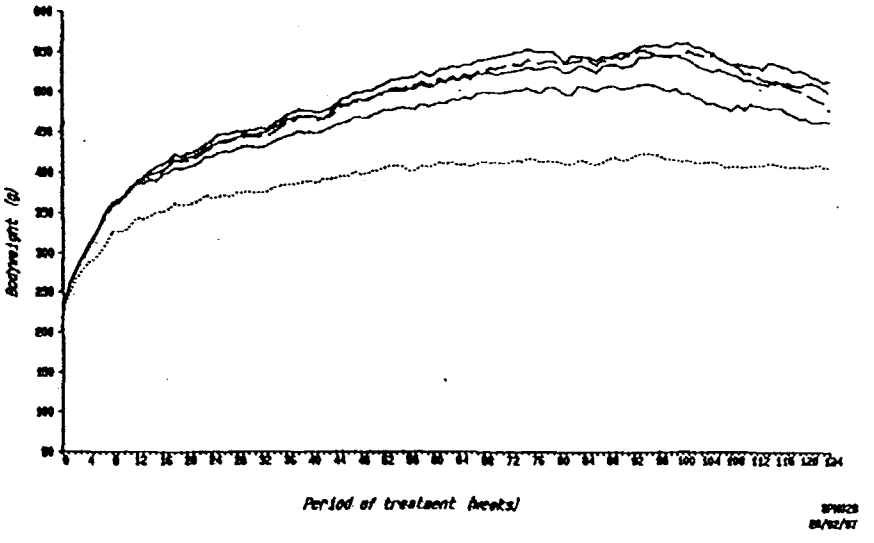
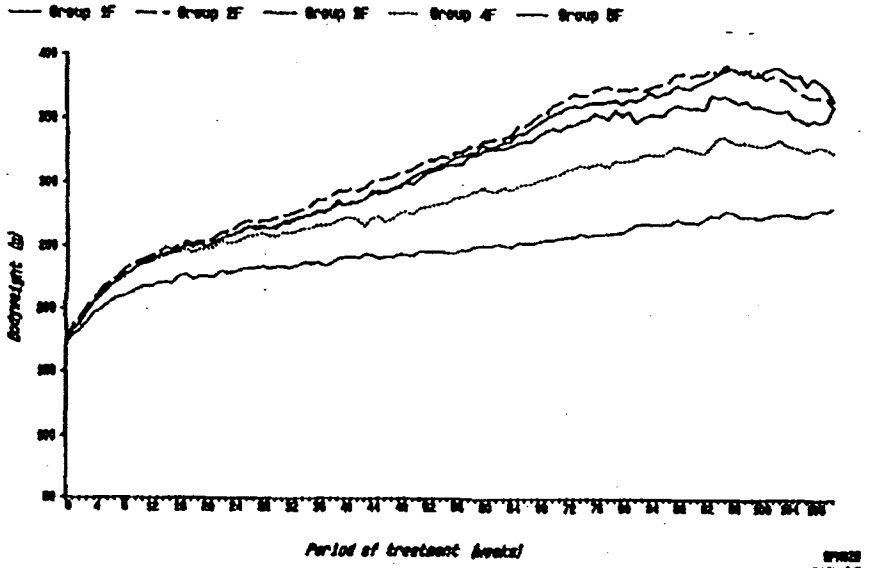


FIGURE 3B
Group mean bodyweight versus period of treatment - females

Group	:	1	2	3	4	5
Compound	:	Control		— SDZ HTF 919 —		
Dosage (mg/kg/day)	:	0	0	20	60	180



4. Food Consumption: Average food consumption in the control group was 20-24 (males) or 17.6-22.1 (females) g/rat/day. The food consumption was ~11-12% lower in the high dose group as compared to the control. The mean achieved doses were within 100-101% of intended doses.

5. Ophthalmology Examination: Higher incidence of posterior capsular and superficial corneal opacity was noted and the information were summarized in a table on page 0029 in volume 18.12 and this table is attached below.

Group incidence (%) of selected ophthalmoscopic findings

	Group and sex				
	1M	2M	3M	4M	5M
Superficial corneal opacity	21	14	23	33	35
	1F	2F	3F	4F	5F
Posterior capsular opacity	44	50	56	73	92

6. Hematology: There were no clear treatment related changes.

7. Gross Pathology: Number of females with pituitary masses was significantly reduced in the mid (30/50) and high (19/50) dose females as compared to the control (39-40/50). Number of females with thickened mammary was significantly reduced in the high dose females (3/50) as compared to the control (10-11/50). Number of females with mammary masses was significantly reduced in the mid (3/50) and high (4/50) dose females as compared to the control (14-15/50). The incidence of cystic ovary was significantly increased in the low (11/50), mid (16/50) and high (18/50) dose females as compared to the control (0-3/50). The incidence of distended uterus was increased in the high dose females (12/50) as compared to the control (3-5/50). The incidence of large or dark mesenteric lymph node was significantly increased in the high dose males (7/50 or 20/50) as compared to the control (1/50 or 5-6/50). This information is summarized in the following table.

Incidence of gross pathological findings

	Con 1	Con 2	Low Dose	Mid Dose	High Dose
# animals with pituitary masses					
Males:	17	15	17	19	15
Females	40	39	33	30	19
Cystic ovary:	3	0	11	16	18
# animals with thickened mammary:					
Males:	0	0	0	1	3
Females:	10	11	12	7	3
# animals with mammary masses:					
Males:	11	5	7	6	0
Females:	15	14	12	3	4
Mesenteric lymph node:					
Dark:					
Males:	5	6	8	9	20
Females:	6	2	2	9	6
Large:					
Males:	1	1	4	1	7
Females:	0	0	0	2	0
Distended uterus:	3	5	4	9	12

Number of animals in each group = 50/sex/group

8. Histopathology:

Non-Neoplastic Changes: The incidence of dilated mammary gland was significantly reduced in the mid (11/50) and high (11/50) dose females as compared to the control (22-24/50). The incidence of reduced follicular development in the ovary was lower in the high dose females (22/50) than that in the control (34-35/50). The incidence of progressive (senile) nephropathy in the kidney was significantly reduced in the high dose males (10/50) as compared to the control (28-33/50). Sponsor believed that these changes were secondary either to the reduced incidence of pituitary tumors or to the lower body weight in these groups. The mucosal hyperplasia in the small intestine was found in 2 control males and 5 high dose animals (4 males and 1 female). This was characterized by increased cellularity, increased basophilia with nuclear crowding in the intestine crypts. In the initial submission, it was found that the incidence of bursal, follicular or luteal cysts was significantly increased in the treatment groups. The incidence of bursal cyst in ovary was 0-2/50, 6/50, 7/50 and 10/50 in the control, low, mid, and high dose groups, respectively (historical control mean: 0.46-0.89%, range: 0-8.2%). The combined incidence of follicular and luteal cysts in ovary was 0-3/50, 9/50, 11/50, and 12/50 in the control, low, mid, and high dose groups, respectively (historical control

mean: 1.6-1.63%, range: 0-10%). This information is summarized in the following table.

Incidence of non-neoplastic histopathological findings

	Con 1	Con 2	Low Dose	Mid Dose	High Dose
Ductular dilation of mammary gland Females:	24	22	24	11	11
Ovary: Reduced follicular development Bursal cyst Combine follicular and luteal cysts	 34 2 3	 35 0 0	 32 6 9	 31 7 11	 22 10 12
Progressive (senile) nephropathy: Males: Females:	 28 23	 33 19	 24 18	 24 19	 10 14
Mucosal hyperplasia in small intestine: Males: Females:	 1 0	 1 0	 0 0	 0 0	 4 1

Number of animals in each group = 50/sex/group

Sponsor was asked to separate the ovarian cysts into bursal, follicular, and luteal cysts and conduct a statistical analysis on each category. In response to this request, sponsor re-evaluated the histopathological preparations of the ovaries from all animals in this study and provided the findings in Table 1 on page 3 of the amendment dated May 25, 2000. This table is attached below.

Table 1. Ovarian pathology in tegaserod treated rats

Dose (mg/kg)	Control 1	Control 2	20 mg/kg	80 mg/kg	180 mg/kg
Number of Animals	50	50	50	50	50
Follicular cysts	15	8	18	12	20
Dilated bursa	1	0	1	7	3
Dialted rete	0	1	0	1	0

The results of statistical analysis indicated that the incidence of follicular cyst was significantly increased in the high dose group as compared to the control 2 ($p = 0.0281$) or the combined control ($p = 0.0465$). Follicular cyst was the only ovarian cysts observed after re-evaluation. Sponsor stated that "the incidence of peer-reviewed histological changes in the ovaries differs from those originally reported because the pathologists panel applied rigorous criteria to diagnose follicular cysts and/or dilated

bursa/rete". However, the detail criteria were not provided. Sponsor also stated that the slides from control and high dose animals in the 6-month rat study were also reviewed and no follicular cysts were observed.

This information was also summarized by sponsor in Table 10F on pages 0207-0225 in volume 18.12. This table is included in Appendix II.

Neoplastic Changes: The incidence of pituitary adenoma was slightly reduced in the high dose females (17/50) as compared to the control (34-35/50). The incidence of this tumor was summarized in a table on page 0032 in volume 18.12 and this table is attached below.

Incidence of pituitary adenoma and focal hyperplasia

Group and sex	1M	2M	3M	4M	5M	1F	2F	3F	4F	5F
Dosage (mg/kg/day)	0	0	20	80	180	0	0	20	80	180
Number examined	49	50	50	50	50	50	50	50	50	50
Pituitary adenoma	16	15	17	19	15	34	35	31	25	17
Focal hyperplasia	8	9	13	8	4	11	8	9	11	19

The incidence of uterine adenocarcinomas was slightly higher in the high dose females (5/50) than in the controls (control = 2 or 3/50, low and mid dose groups = 2/50) and this difference was not statistically significant. Sponsor stated that this incidence (10%) is within the background rate (39%) in female HanIbm Wistar rats (Deerberg, F. Et. Al., Mech. Ageing. Dev., 14:333, 1980). The incidence of benign uterine polyp was slightly but significantly increased in the mid dose group (9/50) as compared to the control group2 (control2 = 2/50, control1, low and high dose = 6/50, 6/50 and 7/50, respectively). There was no evidence for a trend. Therefore, these are not considered treatment related.

There was evidence of statistically significant increase in the following tumors in the high dose group of one sex or the other: benign adrenal cortical adenoma, malignant liposarcoma in kidney and malignant follicular cell carcinoma in thyroid. The incidences of these tumors are summarized in the following table.

No. animals/group = 50	Males					Females				
	con1	con2	low	mid	high	con1	con2	low	mid	high
Adrenal cortical adenoma	1	0	0	1	0	2	0	0	1	3*ab
Liposarcoma in kidney	0	0	0	0	2*a	0	0	0	0	0
Follicular cell carcinoma in thyroid	2	1	0	0	0	0	1	1	0	3*ac

Con1=control1, con2=control2, low, mid and high = 20, 80 and 180 mg/kg/day, respectively
* = p <0.05, a = trend test, b = compared with control 2, c = compared with control 1

The significant increases in the incidences of benign adrenal cortical adenoma, liposarcoma in kidney and follicular cell carcinoma in thyroid were only found in the high dose animals. There were some background incidences of adrenal cortical adenoma and follicular cell carcinoma in the thyroid in control animals and the significant increases in these tumor incidences were only seen in the high dose females when compared with one of the control groups. Liposarcoma in kidney was only found in two high dose males. Therefore, these are not considered treatment related. The incidence of neoplastic histopathological findings extracted from sponsor's table 10C on pages 0170-0175 in volume 18.12 is attached in Appendix II.

9. Plasma Levels: There was no apparent accumulation of the test drug over time. The plasma level of the test drug was slightly lower in the females than in males. These results were summarized in tables 1, 2, 3, 4, 5 and 6 on pages 2388-2393 in volume 18.12. These tables are attached below.

Plasma concentrations of SDZ HTF 919 in rat during a lifespan

oral (diet) oncogenicity study Week 4

Gender	M	F	M	F	M	F
Dose mg/kg/day	20	20	80	80	160	160
Time (h)	Plasma concentrations (ng/ml) : mean ± SD					
0	2.27 ± 0.37	2.04 ± 0.75	28.14 ± 8.92	16.34 ± 3.43	134.65 ± 2.77	125.08 ± 16.19
6	2.17 ± 0.79	1.76 ± 0.61	20.46 ± 3.41	10.27 ± 2.66	86.93 ± 11.43	70.20 ± 16.14
12	1.71 ± 0.27	1.87 ± 0.74	16.39 ± 9.35	6.29 ± 2.34	68.86 ± 12.90	40.44 ± 9.28
16	2.84 ± 1.06	4.83 ± 6.30	17.69 ± 1.39	12.96 ± 3.10	82.02 ± 25.22	79.19 ± 13.36
AUC [0-24h] (ng.h/ml)	54.59	59.96	508.06	275.18	2234.82	1869.40
AUC [0-24h]/Dose (ng.h/ml)/(mg/kg)	2.73	3.00	6.35	3.44	12.42	10.50

Re : Tables A2, A3, A4

Table 1

Plasma concentrations of SDZ HTF 919 in rat during a lifespan

oral (diet) oncogenicity study Week 48

Gender	M	F	M	F	M	F
Dose mg/kg/day	20	20	80	80	160	160
Time (h)	Plasma concentrations (ng/ml) : mean ± SD					
0	6.30 ± 3.48	1.64 ± 0.41	77.93 ± 20.90	43.41 ± 16.87	185.57 ± 32.79	172.63 ± 48.11
6	3.16 ± 0.67	5.09 ± 3.53	80.49 ± 6.42	44.83 ± 19.75	192.66 ± 35.25	136.28 ± 16.89
12	4.85 ± 1.86	1.75 ± 0.51	74.19 ± 15.33	21.13 ± 9.28	156.55 ± 14.28	107.64 ± 26.01
16	3.99 ± 1.14	1.81 ± 0.65	42.52 ± 8.94	13.70 ± 3.73	117.64 ± 36.72	56.60 ± 11.14
AUC [0-24h] (ng.h/ml)	109.60	61.10	1650.78	739.02	3915.61	2850.26
AUC [0-24h]/Dose (ng.h/ml)/(mg/kg)	5.49	3.05	20.63	9.24	21.75	15.83

Re : Tables A6, A7, A8

Table 2