RAT

COVERSHEET FOR CARCINOGENICITY STUDY IN RAT

1. Study No.: G89024 (Report # 308E/92)
2. Name of Laboratory: [redacted]
3. Strain: Sprague-Dawley
4. No./sex/group: 70
5. Doses (O, L, M, H): 0, 0.5, 5, 50 and 200 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:

	Male	Female
O	33/Malignant Sarcoma (skin)	55/Fibroadenoma (mammary gland)
L	31/Benign Adenoma (pituitary)	38/Myelomonocytic Leukemia (spleen)
M (I)	65/Benign Adenoma (pituitary)	38/Myelomonocytic Leukemia (spleen)
M (II)	48/Benign Adenoma (pituitary)	52/Benign Adenoma (pituitary)
H	51/Benign Adenoma (pituitary)	49/Malignant Medulloblastoma (brain)

10. No. alive at termination:

	Male	% Survival	Female	% Survival
O	11/70	15.7	22/70	31.4
L	10/70	14.3	22/70	31.4
M (I)	8/70	11.4	21/70	30.0
M (II)	12/70	17.1	28/70	40.0
H	3/70	4.3	25/70	35.7

11. Statistical Methods Used: Peto etal (International Agency for Research on Cancer Monograph 1980 (Suppl. 2); 311-426)
12. Attach tumor and non-tumor data for each tissue (i.e., benign; malignant; hyperplastic); See Appendix 2

Two-Year Oral (Gavage) Carcinogenicity Study in Sprague Dawley Rats (GTR-31282).

Testing Laboratories: [redacted]

Date Started: October 3, 1989

Date Completed: January 13, 1993

GLP Requirements: A Statement of Compliance with GLP regulations was included.

Testing Species & Strain: Male and Female Sprague-Dawley Rats (8 weeks old, 169-352 g).

No. of Animals: 70/sex/group.

Route of Administration: Oral (gavage).

Dose Levels: 0, 0.5, 5, 50 and 200 mg/kg/day.

Drug Batch No.: 299155/89 PD 324, 399-175/89 PD 341/1 and 399-175/89 PD 341/2.

Methods: In this study dose selection was based on 6-month oral toxicity study in rats in which doses of 0.8, 4, 16 and 320 mg/kg/day were used. The highest tested dose produced significant toxicities (decreased body weight gain, decreased food consumption, microscopic changes in stomach, liver and thyroid etc.). Sponsor selected 0.5, 5, 50 and 200 mg/kg/day levels for the carcinogenicity study in Sprague Dawley rats. Hence, in the present study, groups of rats (70/sex/group) were given orally (gavage) —— 96022-Z at daily doses of 0.5, 5, 50 and 200 mg/kg/day for 2 years. The control group animals received the vehicle (purified water) in similar fashion. The volume of administration was 10 mL/kg and the drug solution pH was adjusted to 10.5 before administration. All animals were observed weekly. Body weights were recorded at pretest, weekly for the first 13 weeks, monthly for the next 42 weeks and weekly thereafter. Food consumption were recorded weekly for the first 13 weeks and then every 3 months thereafter. Blood samples were collected from the vena cava of all surviving rats at termination for hematological tests. Additionally, blood samples were also collected from 5 rats/sex/group at 1 hour after the last dose to measure drug and its thiol metabolite —— 97165) levels in plasma. All surviving rats were sacrificed at the end of study period and subjected to complete necropsy. Only control and high dose group (200 mg/kg/day) animals were examined histologically. Additionally, kidneys, liver, ovaries, sternebrae, thyroid, testes with epididymides and all macroscopic lesions from every other groups were also examined microscopically. Selected specimens of liver and stomach were also examined under electron microscope. Statistical evaluation of neoplastic lesions was performed according to Peto et al. (International Agency for Research on Cancer, Monograph 1980 [Suppl. 2, 311-426]).

Results:

1. **Observed Effects:** Only salivation was seen just after drug administration in treated rats.

2. **Mortality:** Survival rates in females were comparable in all groups. However, at termination, the survival rate in high dose-treated males was significantly lower than the control (16% vs 4%).

Intercurrent Mortality Rates

Male Rats

Days	I	%	II	%	III	%	IV	%	V	%
1-365	2/70	2.8	3/70	4.3	4/70	5.7	7/70	10.0	2/70	2.8
366-545	16/68	23.5	14/67	20.9	16/66	24.2	20/63	31.7	22/68	32.3
546-635	17/52	32.7	28/53	52.8	23/50	46	14/43	32.5	22/46	47.8
636-737	24/35	68.6	15/25	60	19/27	70.4	17/29	58.6	21/24	87.5
Terminal	11	--	10	--	8	--	12	--	3	--
Survival rate	--	15.7	--	14.3	--	11.4	--	17.1	--	4.3

Female Rats

1-365	1/70	1.4	3/70	4.3	2/70	2.8	1/70	1.4	2/70	2.8
366-545	16/69	23.2	10/67	14.9	16/68	23.5	9/69	13.0	14/68	20.6
546-635	15/53	28.3	19/52	33.3	16/52	30.8	17/60	28.3	17/54	31.5
636-737	16/38	42.1	16/38	42.1	15/36	41.7	15/43	34.9	12/37	32.4
Terminal	22	--	22	--	21	--	28	--	25	--
Survival rate	--	31.4	--	31.4	--	30.0	--	40.0	--	35.7

I = Vehicle control
II = 0.5 mg/kg
III = 5 mg/kg
IV = 50 mg/kg
V = 200 mg/kg

It should be noted here that rate of survival was poor in all groups of rats (including control groups) and reached an unacceptable level of less than 50% at termination in control groups and less than 20 males/treatment group were available at termination for analysis. Additionally, from day 526 onward, male rats lost their body weight (including control rats) even though food intakes were not affected by treatment in rats. Sponsor did mention that rats were virus-antibody-free at pretest, however, no documentation was available that rats were screened for infection during the study period. Furthermore, during this period significant increased mortality was seen in high dose treated males when compared to control values. Therefore, significant decrease in survival rate in high dose group could be due to drug as well as possible infection.

3. <u>Body Weight/Food Consumption/Water Consumption</u>: During the first 18 months, all rats gained weight. However, by the end of 526 days the body weight gains were reduced by 9%, 16%, 18% and 21% in 0.5. 5.0, 50 and 200 mg/kg/day treated males respectively, when compared to the control values. It should be noted here that from day 526 onward, male rats (including control) lost their weight. During days 526-722 male rats lost their weights by 22%, 27%, 19%, 16% and 18% at 0, 0.5, 5, 50 and 200 mg/kg/day, respectively. This weight loss during the last 6-months of the study was not evident in female rats. At the end of treatment period (day 722), body weight gains were reduced by 5% and 16% in 50 and 200 mg/kg/day treated females, respectively, when compared to control values. No adverse effects on food intakes were evident. Final body weights for male rats at doses of 0.5, 5, 50, and 200 mg/kg/day were 87.8, 91.4, 94, and 89.1% of the control (764 g) respectively. Final body weights for female rats at doses of 0.5, 5, 50, and 200 mg/kg/day were 109.6, 102.6, 96.5, and 89.1% of the control (576 g) respectively.

Body Weight (g) of Male Rats

Days	Dose (mg/kg/day)				
	0	0.5	5	50	200
6	335 ± 3	331 ± 3	330 ± 3	328 ± 3	327 ± 3
90	626 ± 7	629 ± 9	614 ± 8	592 ± 7	593 ± 6
174	744 ± 10	752 ± 12	728 ± 11	705 ± 9	702 ± 8
265	829 ± 12	831 ± 15	803 ± 13	781 ± 11	775 ± 10
356	889 ± 14	887 ± 16	841 ± 14	819 ± 12	815 ± 11
421	924 ± 15	917 ± 18	857 ± 16	832 ± 15	827 ± 14
526	974 ± 19	915 ± 20	865 ± 18	850 ± 15	829 ± 15
589	918 ± 24	863 ± 27	804 ± 23	829 ± 18	800 ± 21
722	764 ± 37	671 ± 57	698 ± 31	718 ± 23	681 ± 32

Food Consumption (g/animal/day) in Male rats

Days	Dose (mg/kg/day)				
	0	0.5	5	50	200
8	30 ± 0	30 ± 0	30 ± 0	29 ± 0	29 ± 0
92	32 ± 0	32 ± 0	31 ± 0	30 ± 0	32 ± 1
183	31 ± 0	32 ± 1	32 ± 1	31 ± 0	33 ± 0
276	32 ± 1	34 ± 1	31 ± 1	32 ± 0	33 ± 0
365	34 ± 1	36 ± 1	35 ± 1	34 ± 1	36 ± 1
457	35 ± 1	36 ± 1	35 ± 1	35 ± 1	39 ± 1
561	34 ± 1	34 ± 1	33 ± 2	35 ± 1	37 ± 2
638	32 ± 2	30 ± 2	32 ± 2	32 ± 2	34 ± 2
729	31 ± 3	33 ± 2	31 ± 4	36 ± 2	39 ± 11

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Body Weight (g) of Female Rats

Days	Dose (mg/kg/day)				
	0	0.5	5	50	200
6	213 ± 2	216 ± 2	215 ± 2	215 ± 2	215 ± 2
90	338 ± 4	345 ± 4	344 ± 4	335 ± 4	330 ± 3
174	392 ± 5	405 ± 6	396 ± 6	383 ± 5	380 ± 5
265	441 ± 7	458 ± 9	442 ± 7	428 ± 7	420 ± 7
356	490 ± 10	501 ± 11	491 ± 8	466 ± 9	462 ± 8
421	512 ± 10	537 ± 12	524 ± 10	491 ± 10	492 ± 9
526	558 ± 11	586 ± 17	581 ± 14	531 ± 10	520 ± 11
589	561 ± 13	608 ± 18	593 ± 18	537 ± 13	526 ± 13
722	576 ± 25	631 ± 24	591 ± 28	556 ± 18	513 ± 25

Food Consumption (g/animal/day) in Female Rats

Days	Dose (mg/kg/day)				
	0	0.5	5	50	200
8	21 ± 0	21 ± 0	21 ± 0	21 ± 0	21 ± 0
92	21 ± 0	21 ± 0	22 ± 0	21 ± 0	23 ± 0
183	21 ± 0	22 ± 0	22 ± 0	21 ± 0	23 ± 0
276	23 ± 0	23 ± 0	23 ± 0	22 ± 0	24 ± 0
365	24 ± 0	24 ± 1	24 ± 1	24 ± 0	25 ± 0
457	23 ± 0	25 ± 1	26 ± 1	25 ± 0	26 ± 0
561	25 ± 1	28 ± 2	27 ± 1	26 ± 1	27 ± 1
638	25 ± 1	26 ± 1	28 ± 1	26 ± 1	27 ± 1
729	25 ± 1	32 ± 1	29 ± 2	28 ± 2	31 ± 2

4. Hematology/Coagulation/Bone Marrow: No treatment related effects were seen.

5. Plasma Levels of —— 96022 (drug) and its Thiol Metabolite —— 97165: Plasma levels of the drug increased with increasing doses. The thiol metabolite was consistently seen in rats treated with 50 and 200 mg/kg/day dose levels. The levels of thiol metabolite was highly variable (i.e., large standard deviations were seen).

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Plasma Levels of —— 96022 (drug) and its Thiol Metabolite —— 97165) at 1 hr After The Last Dose			
Dose (mg/kg/day)	Sex (M/F)	—— 96022 mcg/ml	——97165 mcg/ml
0.5	M	0.124 ± 0.018	ND
	F	0.152 ± 0.047	ND
5.0	M	0.536 ± 0.16	ND
	F	1.81 ± 1.71	0.063*
50	M	13.1 ± 3.55	0.392 ± 0.15
	F	25.0 ± 9.82	0.604 ± 0.21
200	M	21.0 ± 8.89	0.717 ± 0.25
	F	27.2 ± 20.0	0.702 ± 0.405

ND = not detected —— , mcg/ml
* = seen in only 1 rat

6. Gross Pathology: Increased incidence of thickened fundus of the stomach, enlarged liver, renal enlargement (bilateral) and enlarged parathyroid glands were seen in treated rats. The incidence were as follows:

# Examined	Sex (M/F)	Control 70	0.5 mg/kg 70	5.0 mg/kg 70	50 mg/kg 70	200 mg/kg 70
Stomach						
Thickened fundus	M	0	2	40	43	26
	F	0	3	33	56	42
Liver						
Enlarged	M	1	4	3	11	14
	F	0	1	2	3	5
Kidney						
Renal Enlargement	M	9	15	19	26	29
	F	3	2	3	6	9
Parathyroid						
Enlarged	M	7	8	10	11	15
	F	0	0	1	2	3

7. Histopathology:

Non-Neoplastic Findings: Histopathological examination revealed abnormal changes in stomach, liver, kidney, thyroid, parathyroid, sternebra and testes. Dose-related increased incidence of hyperplasia of the fundic mucosa of the stomach along with increased mucosal height, fundic gland ectasia, presence of eosinophilic chief cells and hyperplasia of ECL cells (chromogranin-positive cells) were seen at 5 mg/kg/day and higher dose levels. Eosinophilic chief cells (fundus) and fundic gland ectasia were also present at the lowest tested dose and none were seen in the controls. Increased incidence of hepatocellular necrosis were seen in high dose treated rats (both sexes). Dose related increased incidence of centrilobular hepatocellular hyperplasia were also seen in treated rats of both sexes. In kidney, dose-related increased incidence of nephropatny were evident in treated rats (both sexes). Fibrous osteodystrophy of sternebra were seen in treated rats. Bilateral parathyroid hyperplasia were seen in treated rats (males: control = 7/68, 0.5 mg/kg/day = 14/56, 5 mg/kg/day = 16/60, 50 mg/kg/day = 14/55 and 200 mg/kg/day = 23/69; females: control = 2/61, 0.5 mg/kg/day = 1/44, 5 mg/kg/day = 3/48. 50 mg/kg/day = 4/55 and 200 mg/kg/day = 5/68). In the thyroid gland, follicular cell hypertrophy was

seen in 50 and 200 mg/kg/day dose group. Thyroid follicular cell hyperplasia was observed in some control as well as treated rats. The incidence for male rats was as follows: control, 1/70; 0.5 mg/kg/day, 0/70; 5 mg/kg/day, 0/70; 50 mg/kg/day, 2/70, and 200 mg/kg/day, 2/70. The incidence for female rats was as follows: control, 2/70; 0.5 mg/kg/day, 0/70; 5 mg/kg/day, 0/70; 50 mg/kg/day, 0/70; and 200 mg/kg/day, 4/70. Polyarteritis nodosa and tubular degeneration were seen in all male rats, but incidences were increased in 50 and 200 mg/kg/day dose groups. Increased incidence of interstitial cell hyperplasia of testes was also evident in high dose group. The incidence of above mentioned abnormalities were as follows:

Number of Rats With Non-neoplastic Findings							
	Sex (M/F)	Control	0.5 mg/kg	5.0 mg/kg	50 mg/kg	200 mg/kg	p-Value Trend Test (FDA)
# Examined		70	70	70	70	70	
Stomach							
Increased height of fundic mucosa'	M	0	0	37	51	36	0.0000
	F	0	0	33	55	53	0.0000
Fundic gland ectasia'	M	0	2	68	62	53	0.0000
	F	0	18	66	66	66	0.0000
Eosinophilic chief cells (fundus)'	M	0	5	53	57	51	0.0000
	F	0	15	59	64	66	0.0000
Hyperplasia, Chromogranin positive cells (fundus)	M	0	0	1	5	3	0.063
	F	0	0	11	20	11	0.0113

Number of Rats With Non-neoplastic Findings (Continued):

Hyperplasia of basal cell, non-glandular	M	1	0	1	3	7	0.0006
	F	1	0	0	0	0	1.0000
Liver							
Eosinophilic cell focus	M	20	21	12	35	47	0.0000
	F	12	7	10	30	36	0.0000
Basophilic cell focus	M	6	8	7	5	6	0.6228
	F	24	29	26	33	31	0.1663
Hepatocellular necrosis	M	11	6	5	10	19	0.0007
	F	7	6	10	12	17	0.0044
Centrilobular hepato-cellular hyperplasia[1]	M	1	5	6	20	39	0.0000
	F	2	4	12	26	38	0.0000
Kidney							
Nephropathy (mild-severe)[1]	M	38	39	47	57	63	0.0000
	F	11	17	14	20	40	0.0000
Sternebra							
Osteodystrophy (fibrous)[1]	M	5	7	11	12	16	0.0086
	F	1	1	0	2	6	0.0019
Testes							
Interstitial cell hyperplasia	M	8	6	5	11	14	0.012
Polyarteritis nodose[1]	M	22	18	23	31	39	0.0001
Tubular degeneration[1]	M	18	18	21	19	33	0.0010
Thyroid							
Follicular cell hypertrophy	M	1	1*	0*	6	20	0.0000
	F	0	0	0	2	5	0.0006

1 = severity of the changes increased with dose
* = only 69 rats were examined

Neoplastic Findings: Drug-related neoplastic findings were evident in the stomach, liver, thyroid gland, kidney, and skin.

Stomach: Treatment with pantoprazole produced benign as well as malignant neuroendocrine cell tumors in dose-related manner from 0.5 to 200 mg/kg/day in female rats. For male rats, gastric carcinoids (benign and malignant) were also seen in 50 and 200 mg/kg/day dose groups. There were 6 benign neuroendocrine cell tumors (found during days 645-734) in 50 mg/kg treated males (8.6%) while only 1 benign neuroendocrine cell tumor (found on day 539) was seen in 200 mg/kg treated males (1.4%). This low incidence rate in high dose treated male was due to decreased survival rates in high dose treated males and tumor (NE cell tumors) tend to appear close to termination of the study. If the survival rate in high dose treated males had been higher, then one would have most likely see a higher incidence rate of neuroendocrine tumors in the stomach of male rats. Additional tumors observed for female rats at 200 mg/kg/day included a malignant adenocarcinoma of chief cell in the fundus (1/70), benign adenomatous polyp in the fundus (1/70), benign squamous cell papilloma in the forestomach (1/70) and malignant squamous cell carcinoma in the forestomach (4/70).

Additional tumors observed for male rats at 200 mg/kg/day included benign adenomatous polyp in fundus (2/70), and benign squamous cell papilloma in the forestomach (7/70) . In rats treated with 50 mg/kg/day, 2/70 females had benign squamous cell papilloma in the forestomach, 2/70 males had malignant squamous cell carcinoma in the forestomach, and 1/70 males also had malignant adenocarcinoma in the pyloric region of the stomach. None of the above findings were seen in control group rats.

Number of Rats With Neoplastic Findings in The Stomach

	Sex (M/F)	Control	0.5 mg/kg	5.0 mg/kg	50 mg/kg	200 mg/kg	p-Value Trend Test (FDA)
# Examined		70	70	70	70	70	
Stomach							
Neuroendocrine cell tumor (benign) fundus	M	0	0	0	6	1	
	F	0	0	4	14	12	0.0001*
Neuroendocrine cell tumor (malignant) fundus	M	0	0	0	1	1	
	F	0	1	2	7	19	0.0000*
Neuroendocrine cell tumor (benign + malignant) fundus	M	0	0	0	7	2	
	F	0	1	6	21	31	0.0000*
Adenocarcinoma of chief cell (malignant) fundus	M	0	0	0	0	0	
	F	0	0	0	0	1	
Adenomatous polyp (benign) fundus	M	0	0	0	0	2	0.0361*
	F	0	0	0	0	1	
Squamous cell papilloma (benign)	M	0	0	0	0	7	0.0000*
	F	0	0	0	2	1	0.1390
Squamous cell carcinoma (malignant)	M	0	0	0	2	0	
	F	0	0	0	0	4	0.0017*
Squamous cell papilloma and carcinoma	M	0	0	0	2	7	0.000*
	F	0	0	0	2	5	0.007*
Adenocarcinoma (malignant) pyloric region	M	0	0	0	1	0	
	F	0	0	0	0	0	---

* = statistically significant

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Liver: Dose related increase incidence of hepatocellular adenomas &/or hepatocellular carcinomas were seen in treated rats.

Number of Rats With Neoplastic Findings in The Liver							
	Sex (M/F)	Control	0.5 mg/kg	5.0 mg/kg	50 mg/kg	200 mg/kg	p-Value Trend Test (FDA)
# Examined		70	70	70	70	70	
Hepatocellular Adenomas (benign)	M	0	2	1	7	9	0.0001*
	F	0	2	0	5	22	0.0000*
Hepatocellular Carcinomas (malignant)	M	1	2	2	0	6	0.0001*
	F	0	1	0	1	3	0.0254*
Hepatocellular Adenoma + Carcinomas	M	1	4	3	7	15	0.0001*
	F	0	3	0	6	25	0.0001*

* = statistically significant

Thyroid: The incidence of follicular cell adenoma was significantly increased in high dose treated rats (both sexes). The incidence of follicular cell carcinoma and C-cell adenomas were also significantly increased in high dose treated females, when compared to the control values.

Number of Rats With Neoplastic Findings in the Thyroid							
	Sex (M/F)	Control	0.5 mg/kg	5.0 mg/kg	50 mg/kg	200 mg/kg	p-Value Trend Test (FDA)
# Examined		70	70	70	70	70	
Follicular cell adenoma	M	1	0	1	1	7	0.0006*
	F	0	0	1	0	3	0.0159*
Follicular cell carcinoma	M	0	1	2	0	1	
	F	1	0	0	0	3	0.0244
Follicular cell adenoma + carcinoma	M	1	1	3	1	8	0.002*
	F	1	0	1	0	6	0.008*
C-cell adenoma	M	3	6	2	3	2	0.5510
	F	3	3	3	2	7	0.0429
C-cell carcinoma	M	0	1	3	0	0	
	F	1	0	1	2	1	
C-cell adenoma + carcinoma	M	3	5	5	3	2	
	F	4	3	4	4	8	

* = statistically significant

Kidney: In kidney, transitional cell carcinoma was seen in 2/70 (p=0.0361, trend test done by FDA) high dose males and none in any other group. Additionally, tubular cell adenoma was seen in 2/70 high dose treated females but none seen in control female rats.

Skin: In high dose treated females, 2/70 (p=0.0318, trend test done by FDA) and 1/70 rats had squamous cell carcinoma of the skin and basal cell carcinoma of the skin respectively, and this finding was not seen in any other groups.

Miscellaneous Findings: Malignant neuroendocrine cell tumors were seen in liver of 1/70 male rats each in 5, 50, and 200 mg/kg/day dose groups. The site of origin for these tumors could not be identified. Malignant neuroendocrine cell tumors were observed in the liver for 1male rat in each the 5 (#4178), 50 (#4883), and 200 (#4997) mg/kg/day groups; however, no histopathological changes were seen in the gastrointestinal tract for these animals. The site of origin of these tumors could not be identified.

In this study, the dose selection was appropriate. At termination, the rate of survival was poor in all groups of rats (including control groups) and reached an unacceptable level of less than 50% in control groups, and less than 20 males/treatment group were available at termination for analysis. It should also be noted that from day 526 onward male rats (including control rats) lost their body weight (16-27%) even though food intakes were not affected by the treatment, No documentation was available that rats were screened for infection during the study period. Furthermore, during this period significant increased mortality was seen in high dose treated males when compared to control values which could be due to drug as well as possible infection. Drug-induced non-neoplastic changes in the stomach included: ECL-cell hyperplasia, increased fundic mucosal thickness, basal cell hyperplasia, eosinophilic chief cells, and fundic gland ectasia. Other non-neoplastic changes included dose-related centrilobular hypertrophy in the liver along with hepatocellular necrosis in high dose group, increased incidence of eosinophilic foci of hepatocellular alteration, thyroid follicular hypertrophy, chronic progressive nephropathy in treated rats (both sexes). In drug-treated males, there was an increased incidence of polyarteritis nodosa in the testis. The neoplastic changes evident in the study were neuroendocrine cell tumor (benign & malignant) in fundus (0%, 1%, 9%, 30% and 44% at 0, 0.5, 5, 50, and 200 mg/kg/day, respectively; p = 0.0000; trend test, H vs C, p=0.0000, Fisher exact test, p values were provided by Dr. Chen [FDA-715]) in treated females. Significant increase in neuroendocrine tumors was not seen in stomach of high dose (200 mg/kg/day) treated males due to decreased survival rates and tumors (NE cell tumors) tend to appear close to termination of the study. If the survival rate in high dose treated males had been higher, then one would have most likely seen a higher incidence rate of neuroendocrine cell tumors in the stomach of male rats. Increased incidence of squamous cell papilloma in stomach was seen in high dose treated males (10% vs 0% in control; p = 0.003 Fisher exact test, calculated by FDA). Increased incidence of squamous cell malignant carcinoma in the stomach was also seen in high dose treated females (5.7% vs 0% in control; p = 0.0017, Fisher exact test [FDA]). According to NTP (report of the NTP Ad Hoc panel on Chemical Carcinogenesis Testing and Evaluation, August 17, 1984, page 275) these two tumors of the forestomach should be combined for analysis purposes. If one combined these tumors (squamous cell papilloma and carcinomas) the results are significant for both sexes (males: control = 0%, low dose = 0%, mid (I) dose = 0%, mid (II) dose = 2.8% and high dose = 10%, p = 0.000, trend test [FDA], females: control = 0%, low dose = 0%, mid (I) dose 0%, mid (II) dose = 2.8% and high dose = 7.1%, p = 0.007, trend test [FDA]). The incidences of squamous cell papilloma and carcinoma in 50 and 200 mg/kg treated rats of both sexes were higher than the incidence in sponsor's historical controls (0.36%, range 0-1.4%). Additionally, chief cell carcinoma in one female (1.4%) and 3 adenomatous polyps (2 females [2.8%] and 1 male [1.4%]) were seen in high dose treated rats. Although incidences of these tumors were not statistically significant by pairwise comparison with the controls, nevertheless these tumors were rare (McMartin et al. Toxicol Pathol 20: 212-225, 1992) and most likely drug-related. Significantly increased incidences of hepatocellular adenomas and carcinoma were seen in treated rats (male incidence: 1%, 6%, 4%, 10% and 21% at 0, 0.5, 5, 50, and 200 mg/kg/day, respectively, [p=0.0001, trend test (FDA); H vs C, p=0.0004, Fisher exact test], female incidence: 0%, 4%, 0%, 9% and 36% at 0, 0.5, 5, 50, and 200 mg/kg/day, respectively, [p=0.0001, trend test (FDA); H vs C, p=0.0000, Fisher exact test). The incidence of hepatocellular adenomas and carcinomas in treated rats (males: 21% and females: 36%) was higher than the incidence in sponsor's historical controls (males: 5%, range 1.4-4.3%; females: 0.7%, range 0-1.4%).

Significantly increased incidences of thyroid follicular cell adenoma plus carcinoma were seen in treated rats (male incidence: 1%, 1%, 4%, 1% and 11% at 0, 0.5, 5, 50, and 200 mg/kg/day, respectively, [p=0.0002, trend test (FDA); H vs C, p=0.0204, Fisher exact test], female incidence: 1%, 0%, 1%, 0% and 9% at 0, 0.5, 5, 50, and 200 mg/kg/day, respectively, [p=0.008, trend test (FDA); H vs C, p=0.0613, Fisher exact test]). The incidence of thyroid follicular cell adenoma plus carcinoma in the high dose treated rats (males: 11% and females: 9%) was higher than the incidence in sponsor's historical controls (males: 2.1%, range 1.4-2.8%; females: 0.7%, range 0-1.4%). Additionally, in kidney, transitional cell carcinoma in high dose treated males (2.8%) and tubular cell adenomas in high dose treated females were seen. It should be noted that 0.5 mg/kg/day treated female (# 4698, killed on day 643) had malignant neuroendocrine cell tumor in the stomach. This study is not acceptable due to significantly decreased survival rate in control rats of both sexes (less than 50% at the termination of the study) and excessive mortality rates in treated males such that only 3-12 rats/group was available for analysis. This decreased survival rate was associated with body weight loss (16-27%) even though food intake was not affected by the treatment, which indicates possible infections. Inspite of these problems, pantoprazole produced tumors in various organs (stomach, liver, thyroid and kidney) in rats of both sexes.

Immunohistological Evaluation of Neuroendocrine Tumor Metastases of Rats Carcinogenicity Study G 89024/TP 1005 (GTR-31279).

Methods: In 2 years carcinogenicity study in SD rats (study G89024, report # 308E/92; 0, 0.5, 5, 50 and 200 mg/kg/day), malignant neuroendocrine cell tumors were seen in liver of 1/70 male rats each in 5 (animal # M4718), 50 (animal # M4883) and 200 (animal # M4997) mg/kg/day dose groups. Additionally, neuroendocrine cell tumors were seen in lymph node (animal # M4870 of 50 mg/kg/day dose group and # F5070 of 200 mg/kg/day) and in multi-organs (abdomen, lung, liver, lymph node and pancreas) of 200 mg/kg/day treated females (# F5108). The site of origin for these tumors were not identified. In the present study, selected slides were stained with hematoxylin Rosin, Grimelius silver stain (for neuroendocrine cells), tyrosine hydroxylase (for evidence of a malignant medullary tumor of the adrenals), glucagon (for evidence of a malignant islet cell tumor of the α-cells of the pancreas) and insulin (for evidence of a malignant islet cell tumor of β-cells of the pancreas), S100 (for perineural Schwann cell) and lipase (for the detection of exocrine cells of the pancreas) for immunohistological evaluation.

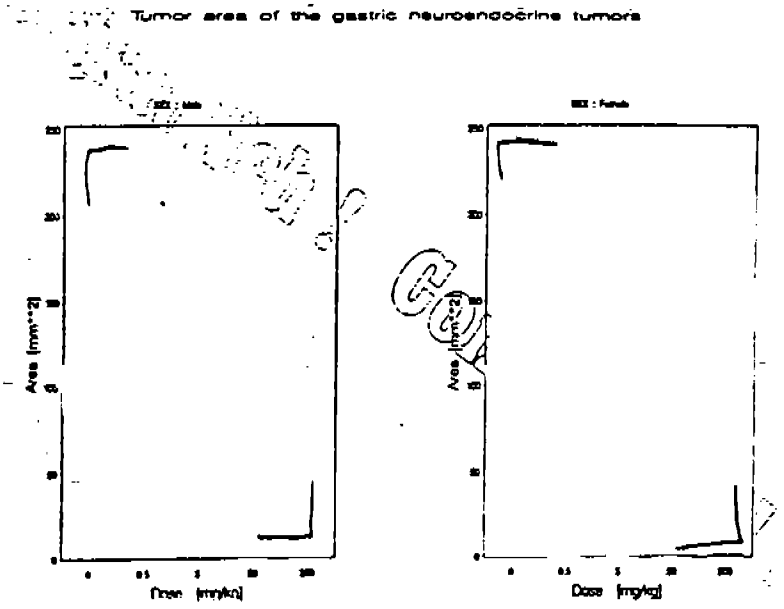
Results: The liver metastases of rats M4718 and M4997 showed marked insulin expression (+++) and the Grimelius reaction was partly positive (+ to ++). Thus, the primary site of tumor in these rats were considered to be pancreas (islet cell carcinoma). The liver metastasis of rat # M4883 was most likely anaplastic sarcoma (Grimelius silver stain and chromogranin stain were negative, therefore, neuroendocrine tumor classification can be excluded). Animal # F5108 gave positive reaction with chromogranin and Grimelius silver staining, thus it had neuroendocrine tumors in the stomach (confirming the previous diagnosis). The tumor of animal # M4870 reacted strongly with monoclonal antibody against lipase, hence it was diagnosed to be acinar cell carcinoma of pancreas. The results of immunohistological staining of the tumor of animal # F5070 was not conclusive. In the

present report (# 58/95 K1) sponsor indicated that the original diagnoses in at least 4 out of 6 rats (# 4178, # 4997, # 4870 and # 4883) were incorrect. According to the new diagnoses, animal # 4178 and # 4997 had islet cell carcinoma of pancreas, animal # 4870 had acinar cell carcinoma of pancreas and animal # 4883 had "most likely" anaplastic sarcoma. It should be noted that in the original histopathology reports of animals # 4178, # 4997 and # 4870 there was no mention of carcinomas in the pancreas.

Number and Size of Gastric Neuroendocrine Tumors in the SD Rat Carcinogenicity Study (GTR-31280).

Methods: In this study chromogranin stained slides of all gastric neuroendocrine tumors (NET) seen in SD rat carcinogenicity study (report # 308E/92, 0, 0.5, 5, 50 and 200 mg/kg/day) were reevaluated and number and size of NET were recorded.

Results: In the earlier submitted report of SD rat carcinogenicity study, incidence of NET (benign + malignant) in males were 0/70, 0/70, 7/70 and 2/70 at 0.5, 5, 50 and 200 mg/kg/day dose levels, and the corresponding incidence in females were 1/70, 6/70, 21/70 and 31/70, respectively (for detail see review dated 8/10/94). In the present reevaluation, sponsor classified NET tumors into 4 sizes. In treated females, irrespective of size of tumor, number of tumor increased with increasing dosages and tumor area also increased with increasing dosages. This reanalysis did not add any new information to the previously submitted results.



Carcinogenicity Study of Pantoprazole in Fischer CDF (F-344) /Crl BR Rats

COVERSHEET FOR CARCINOGENICITY STUDY IN RAT

1. Study No.: KR0143 (Report # 220/94)

2. Name of Laboratory: Byk Gulden
Hamburg, Germany

3. Strain: Fischer CDF (F-344)/Crl BR

4. No./sex/group: 50

5. Doses (0,0, L, M, H): 0, 0, 5, 15 and 50 mg/kg/day

6. Basis for dose selection stated: No

7. Interim sacrifice: No

8. Total duration (weeks): 110

9. Week/site for first tumor: Chronological listing of tumor findings was not provided (Day of Mortality and Presumed Cause of Death).

	Male	Female
Untreated	Malignant lymphoma, day 656	Squamous cell carcinoma of oral cavity, day 624
0	Lymphoma/leukemia, day 479	Lymphoma/leukemia, day 521
L	Lymphoma/leukemia, day 545	Pituitary adenoma, day 247
M	Liposarcoma, day 257	Malignant lymphoma, day 313
H	Malignant lymphoma, day 425	Squamous cell carcinoma of the oral cavity infiltrating the Harder's gland and nasal cavity, day 522

10. No. alive at termination:

	Male	% Survival	Female	% Survival
0	27/50	54	29/50	58
0	25/50	50	17/50	34
L	20/50	40	13/50	26
M	28/50	56	15/50	30
H	22/50	44	15/50	30

11. Statistical Methods Used: Peto et al. (International Agency for Research on Cancer Monograph 1980 (Suppl. 2]; 311-426)

12. Attach tumor and non-tumor data for each tissue (i.e., benign; malignant; hyperplastic): See Appendix 1

RODENT CARCINOGENICITY FACTSHEET

NDA: **IND:** —— **DRUG CODE:**
CAS #: **DATE:**
DIVISION(s): HFD-180
DRUG NAME(s): Pantoprazole

SPONSOR: Wyeth-Ayerst Laboratories
Philadelphia, PA

LABORATORY:

P/T Reviewer(s): Tanveer Ahmad, Ph.D.
P/T REVIEW DATE: June 28, 1996
CARCINOGENICITY STUDY REPORT DATE: April 4, 1996
THERAPEUTIC CATEGORY:

PHARMACOLOGICAL/CHEMICAL CLASSIFICATION: Proton Pump Inhibitor

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

MUTAGENIC/GENOTOXIC (Y/N/equivocal/na; assay): Pantoprazole was clastogenic in cultured human lymphocytes. In the presence of S-9 mix it gave a positive result in CHO/HGPRT forward gene mutation assay. Equivocal results were seen in mouse micronucleus tests (first test was positive while second test was negative). In ex-vivo liver DNA binding study, drug or its metabolize binds to rat liver DNA.

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

RAT STUDY DURATION (weeks): 104 STUDY
STARTING DATE: July 20, 1992
STUDY ENDING DATE: April 4, 1996
RAT STRAIN: F-344
ROUTE: Oral (gavage)
DOSING COMMENTS: Highest tested dose is not the maximum tolerated dose (MTD).

No. Rats in Control1 (C1): 50 **Control2 (C2):** 50
Low Dose (LD): 50 **Middle Dose (MD):** 50
High Dose (HD): 50 **High Dose2 (HD2):**

RAT DOSE LEVELS (mg/kg/day)

Rat Low Dose: 5 **Rat Middle Dose:** 15
Rat High Dose: 50 **Rat High Dose2:**

Basis for Doses Selected (MTD; AUC ratio; saturation; maximum feasible): Not given.

RAT CARCINOGENICITY (negative; positive; MF; M; F.): Positive

RAT TUMOR FINDINGS: Benign and malignant gastric carcinoids.

RAT STUDY COMMENTS: In the 2-year oral (gavage) carcinogenicity study in Fischer F-344 rats (0, 0, 5, 15 and 50 mg/kg/day), dose selection was based on the results of earlier submitted carcinogenicity study in Sprague Dawley rats (study # G89024: 0, 0.5, 5, 50 and 200 mg/ kg/day via gavage) in which, according to sponsor, the highest tested dose (200 mg/kg/day) far exceeded the MTD (based on low survival rate at termination in all groups [including controls: 4-17% in males and 30-40% in females] and body weight loss of 16-27% [including controls] during the last six month of the study in males). Sponsor's conclusion that a dose of 200 mg/kg/day far exceeded the MTD in SD rats is not correct because in SD rat carcinogenicity study, survival rate was excellent up to 18-month of treatment (61-76% in males and 74-86% in females) and body weights in 0.5, 5, 50, and 200 mg/kg/ day treated males were 6%, 11%, 13%, and 15%, lower than the control, while body weights in females treated with 50 and 200 mg/kg/day were 5% and 7% lower than control respectively. Hence, 200 mg/kg/day did not exceed the MTD in SD rats (for detail see review dated 8/10/94). Furthermore, it is not proper to assess MTD in one strain of rat and conduct carcinogenicity study in another strain of rat. Sponsor either should have repeated carcinogenicity study in Sprague Dawley rat or should have conducted a 3-month dose ranging study in Fischer F-344 rats before conducting the carcinogenicity study in Fischer F-344 rats. Hence, dose selection in the present study is not appropriate. Treatment had no significant effect on mortality rates, body weights and food consumption. Highest tested dose in the present study is not MTD. In spite of these deficiencies, pantoprazole induced gastric carcinoids (benign as well as malignant) in treated males (mid and high dose groups) and females (all treated groups) and none in the control rats (cage control or vehicle control). In Sprague Dawley rats, pantoprazole produced tumors not only in stomach, but it also produced tumor in liver and thyroid and nephropathy in kidneys. If MTD would have been used in this study, then one would most likely see higher rate of incidence of neuroendocrine cell tumors and probably other tumors in different organs (liver, kidney, adrenals etc). Present study is not very informative, it certainly confirms some of the findings of the earlier reported carcinogenicity study in Sprague Dawley rats.

Two-Year Oral (gavage) Carcinogenicity Study in Fischer 344 Rats (GTR-31898 and GTR-31545).

Testing Laboratories: Byk Gulden
Hamburg, Germany

Date Started: July 20, 1992

Date Completed: April 4, 1996

GLP Requirements: A Statement of Compliance with GLP regulations was included.

Testing Species & Strain: Male and Female Fischer CDF (F-344)/ Crl BR Rats (6 weeks old, females: 69-131 g and males: 72-103 g).

No. of Animals : 50/sex/group.

Route of Administration: Oral (gavage).

Dose Levels: 0, 0, 5, 15 and 50 mg/kg/day.

Drug Batch No.: 500-205

Methods: Sponsor earlier submitted results of carcinogenicity study in Sprague-Dawley rats. In this study, survival rate at termination in all groups (including controls) were very low (4-17% in males and 30-40% in females). Furthermore, decreased survival rate in males, was associated with body weight loss of 16-27% (including controls) during the last six month of the study. In spite of these problems, pantoprazole produced tumors in various organs (stomach, liver, thyroid and kidney) in rats of both sexes. Sponsor concluded that the highest tested dose (200 mg/kg/day) far exceeded the MTD (in SD rats), therefore, they repeated the carcinogenicity study in Fischer F-344 rats and arbitrarily selected 5, 15 and 50 my/kg/day dose levels. Sponsor's conclusion that a dose of 200 mg/kg/day far exceeded the MTD in SD rats is not correct because in SD rat carcinogenicity study, survival rate was excellent up to 48-month of treatment (61-76% in males and 74-86% in females) and body weights in 0.5, 5, 50 and 200 mg/kg/day treated males were 6%, 11%, 13% and 15% lower than the control, while body weights in females treated with 50 and 200 mg/kg/day were 5% and 7% lower than control, respectively. Hence, 200 mg/kg/day did not exceed the MTD in SD rats. In the present study, groups of rats (50/sex/group) were given orally (gavage) pantoprazole at daily doses of 5, 15 and 50 mg/kg/day for 2 years. There were 2 control groups, one group was used as cage control while the other group received the vehicle (purified water, pH 10.5) in similar fashion. The volume of administration was 10 mL/kg and the drug solution pH was adjusted to 10.5 before administration. All animals were observed once/twice daily. Body weights were recorded at pretest, weekly for the first 14 weeks, monthly thereafter. Food consumptions were recorded weekly for the first 14 weeks and then every 4 weeks thereafter. Blood samples were collected from the retro-orbital venous plexus of all surviving rats at termination for hematological tests. Additionally, blood samples were also collected from 6 rats/sex/treatment group at 0.25, 0.5, 1, 2, 3 and 5 hour after the first, 367th and 710th dose to measure drug and its thiol metabolite (—— 97165) levels in plasma (day 1 samples were misplaced and never analyzed). All surviving rats were sacrificed at the end of study period and subjected to complete necropsy histological examinations. Statistical evaluation of neoplastic lesions was performed according to Peto et al. (International Agency for Research on Cancer, Monograph 1980 [Suppl. 2, 311-426]).

Results:

1. Observed Effects: No treatment related effects were seen.

2. Mortality: Treatment had no significant effect on intercurrent mortality rates (see below) At termination survival rates were comparable in all groups (40-56% in males and 26-34% [survival in cage control females was 58%] in females).

Intercurrent Mortality Rates

Male Rats

Weeks	I	%	II	%	III	%	IV	%	V	%
1-52	1/50	2.0	5/50	10.0	7/50	14.0	2/50	4.0	4/50	8.0
53-78	2/49	4.1	5/45	11.1	2/43	4.6	4/48	8.3	5/46	10.9
79-110	20/47	42.5	15/40	37.5	21/41	51.2	16/44	36.4	19/41	46.3
Terminal	27	--	25	--	20	--	28	--	22	--
Survival rate	--	54	--	50	--	40	--	56	--	44

Female Rats

1-52	8/50	16.0	18/50	36.0	21/50	42.0	21/50	42.0	21/50	42.0
53-78	2/42	4.8	7/32	21.8	7/29	24.1	3/29	10.3	8/29	27.6
79-110	11/40	27.5	8/25	32.0	9/22	40.9	11/26	42.3	6/21	28.6
Terminal	29	--	17	--	13	--	15	--	15	--
Survival rate	--	58	--	34	--	26	--	30	--	30

I = Cage control
II = Vehicle control
III = 5 mg/kg
IV = 15 mg/kg
V = 50 mg/kg

It should be noted here that rate of survival was poor in all groups of female rats (including vehicle control group and excluding cage) and reached an unacceptable level of less than 50% at termination. In vehicle control and treated control groups, less than 20 females/ group were available at termination for analysis.

3. Body Weight/Food Consumption/Water Consumption: Treatment had no significant effect on body weight and food consumptions, when compared to the vehicle control values.

MEAN BODY WEIGHT (g) OF MALE RATS					
Weeks	Control 1	Control 2	Low Dose	Mid Dose	High Dose
1	107	107	106	106	108
13	263	250	251	248	250
54	378	355	353	354	362
105	352	360	325	350	351
MEAN BODY WEIGHT (g) OF FEMALE RATS					
1	89	88	87	88	88
13	151	143	138	144	140
54	192	182	177	181	178
105	237	212	202	203	203

FOOD CONSUMPTION (g/animal/day) IN MALE RATS					
Weeks	Control 1	Control 2	Low Dose	Mid Dose	High Dose
1	15.9	15.6	15.5	15.6	15.7
13	17.6	16.9	17.0	17.3	17.5
50-53	18.3	17.5	17.6	17.8	18.2
102-104	14.8	17.6	16.4	16.7	17.5
FOOD CONSUMPTION (g/kg/day) IN FEMALE RATS					
1	12.9	12.5	12.2	12.5	12.4
13	11.2	10.3	10.0	10.5	10.2
50-53	11.4	11.2	11.1	11.3	10.9
102-104	13.6	12.6	12.4	12.4	11.9

4. Hematology/Coagulation/Bone Marrow: No treatment-related effects were seen.

5. Gross Pathology: Increased incidence of thickened gastric glandular mucosa were seen in treated rats (males: cage control = 3/27 [11.1%], vehicle control = 5/25 [20%], low dose = 16/20 [80%], mid dose = 18/28 [64.3%] and high dose = 15/22 [68.2%]; females: cage control = 2/29 [6.9%], vehicle control = 0/17 [0%], low dose = 6/13 [46.1%], mid dose = 7/15 [46.6%] and high dose = 7/15 [46.6%].

6. Histopathology:

Non-Neoplastic Findings: Histopathological examination revealed abnormal changes in liver, kidney, adrenals and stomach.

In liver, increased incidence of pigment deposits in high dose treated males [cage control = 2/50 (4%), vehicle control = 4/50 (8%), low dose = 8/50 (16%), mid dose = 7/50 (14%) and high dose = 8/50 (16%)] and increased incidence of spongiosis in mid and high dose treated males [cage control = 10/50 (20%), vehicle control = 9/50 (18%), low dose = 9/50 (18%), mid dose = 9/50 (18%) and high dose = 15/50 (30%)] were seen. Increased incidence of centrilobular hepatocellular hypertrophy were also seen in treated rats of both sexes (see below).

In kidney, dose related increased incidence of interstitial nephritis were seen in treated males [cage control = 0/50 (0%), vehicle control = 1/50 (2%), low dose = 2/50 (4%), mid dose = 3/50 (6%) and high dose = 4/50 (8%)]. Chronic progressive nephropathy was seen in all rats (including control groups), but the severity was increased in high dose treated male rats. In adrenals, increased incidence of pigment deposits in mid and high dose treated males [cage control = 17/50 (34%), vehicle control = 16/50 (32%), low dose = 37/50 (34%), mid dose = 25/50 (50%) and high dose = 35/50 (70%)] were also seen.

In forestomach, dose related increased incidence of hyperplasia of the squamous epithelium were seen in treated rats. In fundus, increased incidences of eosinophilic chief cells hyperplasia, glandular ectasia and basal fibrosis were seen in treated rats. Significant increase in Grimelius positive cells hyperplasia (focal and chain) in the fundus were seen in treated rats of both sexes (micronodules were seen in mid and high dose treated rats). Significant increase in mucosal hyperplasia of antrum was seen in mid and high dose treated rats.

The incidence of above mentioned abnormalities were as follows:

Number of Rats With Non-neoplastic Findings							
# Examined	Sex (M/F)	CC	VC	5 mg/kg	15 mg/kg	50 mg/kg	p-Value Trend Test
		50	50	50	50	50	
Stomach							
Hyperplasia of squamous epithelium (forestomach)	M	2	1	2	4	8	
	F	3	1	0	1	4	
Eosinophilic chief cells hyperplasia (fundus)	M	0	0	34	40	39	
	F	0	0	40	42	45	
Glandular ectasia (fundus)	M	17	16	36	42	44	
	F	23	16	40	39	29	
Basal fibrosis (fundus)	M	11	7	20	26	25	
	F	1	0	5	16	11	
Liver							
Centrilobular hepato-cellular hypertrophy	M	1	13	12	19	14	
	F	1	10	10	23	26	

Number of Rats With Non-Neoplastic Findings							
	Sex (M/F)	CC	VC	5 mg/kg	15 mg/kg	50 mg/kg	p-Value Trend Test
Stomach							
Grimelius positive cells focal hyperplasia (funds)	M	0/39	0/32	4/32	3/34	2/30	
	F	0/35	1/23	4/17	4/17	2/20	
Grimelius positive cells chain (funds)	M	2/39	0/31	14/32	15/34	12/30	
	F	0/35	0/23	10/17	11/25	11/20	
Grimelius positive cells micronodules (funds)	M	0/39	0/32	0/32	2/34	3/30	
	F	0/35	0/23	0/17	2/25	4/20	
Mucosal Hyperplasia (antrum)	M	0/49	9/49	6/50	12/50	19/50	
	F	2/49	1/50	4/46	15/49	12/47	

Neoplastic Findings: Increased incidence of benign as well as malignant neuroendocrine cell tumors (gastric carcinoids) were seen in treated rats (males: cage control = 0%, vehicle control = 0%, low dose = 0%, mid dose = 8% and high dose = 14%; females: cage control = 0%, vehicle control = 0%, low dose = 8%, mid dose = 24% and high dose = 14%).

Number of Rats With Neoplastic Findings In The Stomach

# Examined	Sex (M/F)	CC	VC	5 mg/kg	15 mg/kg	50 mg/kg	p-Value Trend Test (FDA)
		50[1]	50[2]	50	50	50	
Stomach							
Neuroendocrine cell tumor (benign) fundus	M	0	0	0	2	5	
	F	0	0	2	9	4	
Neuroendocrine cell tumor (malignant) fundus	M	0	0	0	2	2	
	F	0	0	2	3	3	
Neuroendocrine cell tumor (benign + malignant) fundus	M	0	0	0	4	7	
	F	0	0	4	12	7	

* = statistically significant
1 = only 49 males
2 = only 49 females

Miscellaneous Findings: In treated males, squamous cell papilloma in the nasal cavity (in 1/50 high dose group), basal cell carcinoma of the skin (in 1/50 high dose group), mammary gland fibroma (in 2/49 high dose group), adenocarcinoma of rete testis (in 1/50 high dose group), meningioma of the brain (in 1/47 high dose group) and granulocytic leukemia (in 1/20 mid dose group and in 2/27 high dose group) were also seen. In treated females, parathyroid gland adenoma was seen in 1/43 high dose group. These tumors were seen only in one sex and incidence rates are "very low", therefore considered not to be treatment related.

In the 2-year oral (gavage) carcinogenicity study in Fischer F-344 rats (0, 0, 5, 15 and 50 mg/kg/day), dose selection was based on the results of earlier submitted carcinogenicity study in Sprague Dawley rats (0, 0.5, 5, 50 and 200 mg/ kg/day via gavage) in which, according to sponsor, the highest tested dose (200 mg/kg/day) far exceeded the MTD based on low survival rate at termination in all groups [including controls: 4-17% in males and 30-40% in females] and body weight loss of 16-27% (including controls) during the last six month of the study in males . Sponsor's conclusion that a dose of 200 mg/kg/day far exceeded the MTD in SD rats is not correct because in SD rat carcinogenicity study, survival rate was excellent up to 18 months of treatment (61-76% in males and 74-86% in females) and body weights in 0.5, 5, 50 and 200 mg/kg/day treated males were 6%, 11%, 13% and 15% lower than the control, while body weights in females treated with 50 and 200 mg/kg/day were 5% and 7% lower than control, respectively. Hence, 200 mg/kg/day did not exceed the MTD in SD rats. Furthermore, it is not proper to assess MTD in one strain of rat and conduct carcinogenicity study in another strain of rat. Sponsor either should have repeated carcinogenicity study in Sprague Dawley rat or should have conducted a 3-month dose ranging study in Fischer F-344 rats before conducting the carcinogenicity study in Fischer F-344 rats. Hence, dose selection in the present study is not appropriate. Treatment had no significant effect on mortality rates, body weights and food consumption. Highest tested dose in the present study is not MTD. Inspite of these

problems, pantoprazole induced gastric carcinoids (benign as well as malignant) in treated males (mid and high dose groups) and females (all treated groups) and none in the control rats (cage control or vehicle control). In Sprague Dawley rats, pantoprazole produced tumors not only in stomach, but it also produced tumors in liver and thyroid and nephropathy in kidneys. If MTD had been used in this study, then one would most likely see higher rate of incidence of neuroendocrine cell tumors and probably other tumors in different organs (liver, kidney, adrenals etc). The present study is not very informative; although, it certainly confirms some of the findings of the earlier reported carcinogenicity study in Sprague Dawley rats.

Addendum:

Maximum Tolerated Dose: In a 90-day oral dose range finding study (GTR-33264, Initiated on July 15, 1992, five days prior to starting the carcinogenicity study with Fischer rats), Fischer 344 rats received pantoprazole at 200 mg/kg/day as a comparator to rats that received B8401-026, the thiol metabolite of pantoprazole, at doses of 20 or 50 mg/kg/day. For rats that received pantoprazole at 200 mg/kg/day, there was no mortality or impairment of body weight gain. For rats that received pantoprazole at 200 mg/kg/day, the target organs of toxicity were the stomach, lungs, liver, and thyroid gland. For the glandular stomach/fundus, histopathological changes included: chief cell hyperplasia, parietal cell degeneration and vacuolation, glandular ectasia, eosinophilic chief cells, inspissated secretory products, parietal cell swelling, submucosal lymphocytic infiltration, and increased mucosal height. For the stomach with Grimelius stain, histopathological changes included: GPC hyperplasia, and GPC hyperplasia with chains. For the lungs, histopathological changes included: round/mixed cell infiltration. For the liver, histopathological changes included centrolobular swelling. For the thyroid gland, histopathological changes included: activation. Pantoprazole at 200 mg/kg/day could be considered a maximum tolerated dose in Fischer 344 rats.

Serum Drug Levels: Serum levels of pantoprazole and its sulfone metabolite were measured on days 1, 367, and 710 in Fischer rats that received pantoprazole at doses of 5, 15, and 50 mg/kg/day. The sulfone of pantoprazole is the main serum metabolite in the rat. Serum AUC values for pantoprazole were generally higher in female rats than male rats on days 1, 367, and 710. In contrast, serum AUC values for the sulfone metabolite were generally higher in male rats than female rats on days 1, 367, and 710. AUC values for pantoprazole on days 1, 367, and 710 increased in a dose proportional manner. Maximum serum concentrations for pantoprazole were observed at the first time point (i.e., T_{max} = 0.25 hr). Pantoprazole was not detected prior to dosing on days 367 or 710 suggesting that there was no accumulation of the parent compound. AUC values on days 367 and 710 were relatively comparable; however, AUC values on day 1 were significantly lower. This difference could be due to the use of different groups of rats as well as a lack of steady state concentrations on day 1. Half-life values for pantoprazole increased in a dose-related manner. Half-life values for the sulfone were longer than those observed for pantoprazole and they displayed no relationship to dose. AUC values for the sulfone increased with ascending doses; however, increases were greater than proportional to dose. AUC values for the sulfone were greatest on day 367. The sulfone was not detected prior to dosing on days 367 or 710 suggesting that there was no accumulation of the metabolite.