CANCELLED FILE
"7-21-83"

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

AUG 12 1983

MEMORANDUM

TO: Henry Jacoby
Product Manager (21)
Registration Division (TS-767)

THRU: Edwin R. Budd, Section Head
Section II, Toxicology Branch
Hazard Evaluation Division (TS-769)

Budd 7/21/83

SUBJECT: EPA Reg. No. 239-1246. Captan. Review of "Two-Year Oral Toxicity/Carcinogenicity Study of Captan in Rats." IRDC; #153-097; June 23, 1982. Submitted by Stauffer Chemical Company and Chevron Chemical Company. EPA Accession No. 249335-249338 and 249731 (Additional Data; dated March 1, 1983).

TOX Chem No. 159

Summary:

Captan was administered in the diet to male and female rats for 24 months at dose levels of 0, 25, 100, and 250 mg/kg/day, adjusted for body weight and food consumption figures recorded biweekly.

Hematology, blood chemistry, urinalyses and ophthalmoscopic studies were performed at 6 month intervals.

Interim necropsies were performed on 10 rats/sex/dose at 12 and 18 months. Gross necropsies and histopathological examinations of all major tissues and organs were performed on all rats that died or were sacrificed during the study.

Captan produced no effects on general appearance, behavior, food consumption, ophthalmology, overall mortality, hematology, blood chemistry or urinalyses. There was a dose-related statistically significant decrease in the mean body weights of both sexes at the mid- and high-dose levels throughout the study.

1

Gross necropsies were negative for macroscopic lesions of toxicological significance. Statistically significant increases in organ weights were seen as follows:

Increases in Organ Weights.

	Dose mg/kg/day	Adrenals		Kidneys		Heart		Brain		Liver		Thyroid/ Parathyroid	
		M	F	M	F	M	F	M	F	M	F	M	F
12 months	25	AR	-	-	-	R	-	R	-	-	-	-	-
	100	R	-	R	-	R	-	R	-	-	-	-	-
	250	R	R	R	R	R	R	R	-	-	-	-	-
18 months	25	-	-	-	-	-	-			-	-	-	-
	100	AR	-	-	-	-	-			-	-	-	-
	250	-	-	AR	-	R	-			AR	AR	-	-
Final sacrifice	25	-	-	-	-	-	-	-	-	-	-	A	-
	100	-	-	AR	R	-	AR	-	R	-	R	-	R
	250	-	-	R	R	R	A	R	R	R	R	R	R

A: Statistically significant increase in absolute weight.
R: Statistically significant increase in relative weight.

Increased hepatocellular hypertrophy was reported in both sexes at the 18-month interim sacrifice at the high dose and at the end of the study for males at the mid and high dose.

No statistically significant treatment related differences in total numbers of neoplastic lesions, total benign neoplasms, total malignant neoplasms, or in the number of specific neoplasms were seen in this study except for kidney tumors in males.

A possible dose related trend was seen for kidney tumors (combined cortical cell adenomas/tubular adenomas and renal cell carcinoma/tubular adenocarcinomas) in the males (1%, 1%, 4%, and 6% at 0, 25, 100, and 250 mg/kg/day).

Recommendations:

1. We need the original copies of the pages that were marked "revised" in this submission, and/or an adequate explanation for the numerous revisions. See p. 15 of this review for further explanation.

2. In order to evaluate the tumors seen we also would like historical rat control data on the incidence of kidney tumors in IRDC Charles River CD rats on a study-by-study basis for

the last several years. These additional data have been already requested through the SPRD Product Manager, Carol Langley.

3. Core classification of this study is reserved until the information requested in 1. and 2. above is received and evaluated.

4. The tentative NOEL for this study is 25 mg/kg/day. The LEL is 100 mg/kg/day.

DETAILED REVIEW OF STUDY

Study Title and Description: 2-Year Oral Toxicity/Carcinogenicity Study in Rats, IRDC, June 23, 1982, was submitted by Stauffer Chemical Company and Chevron Chemical Company.

Identification: 5 Volumes of EPA Accession Numbers: 249338, 249335, 249336, 249337 and 249731 (additional data dated March 1, 1983). IRDC Laboratory study number: 153-097

Sponsor: Chevron Chemical Company
Stauffer Chemical Company

Laboratory: International Research and Development Corporation (IRDC) Mattawan, Michigan 49071

Test Material: Captan Technical, SX 944, 89% pure.
Jugs No. 1-9, 3/24/78
Jugs No. 10 3/24/78

STUDY METHODS

Animals:

Male and female CD rats, from the Charles River Breeding Laboratories, Portage, Michigan were four weeks old at initiation of study.

Dosage Groups:

Males and females were randomly assigned (by computer generated random numbers) to the following groups:

Dose Level (mg/kg/day)	Male Rats	Female Rats
0	70	70
25	70	70
100	70	70
250	70	70

One male rat from a reserve group (5 male and 5 female rats at each dose level) was used to replace a male rat that died at day 26 of the experiment. No other reserve group animals were used.

The diet was administered for 24 months from October 3, 1978 to terminal sacrifice on October 1 to 3, 1980.

Animal Maintenance:

The rats were caged individually and the humidity, temperature and light (12 hours on) were controlled. Water and diets were available ad libitum.

Test Diet:

Fresh diets of captan added to commercially prepared laboratory rodent feed were made up weekly. The concentrations of captan were adjusted using the most recent body weight and food consumption figures, measured weekly for the first 14 weeks and once every two weeks after that. Water consumption was not recorded.

Observations:

Observations for signs of toxicity, moribundity and mortality were made twice daily. A detailed examination was done each week. Food consumption and individual body weights were recorded weekly for the first 14 weeks and once every 2 weeks for the remainder of the study.

Ophthalmoscopic examinations were performed on all animals at 0, 6, 12, 18 and 24 months. Ten rats/sex at 0 months and ten rats/sex/group at 6, 12 and 18 months were also randomly selected for clinical blood testing and a fasting urinanalysis. Twenty rats/sex/group were selected at 24 months. The following hematology and blood chemistry studies were performed:

Erythrocyte count
Hemoglobin
Hematocrit
Leucocyte count
Prothrombin time

Activated partial thromboplastin time
Platelet count
Glucose
Blood urea nitrogen
Alkaline phosphatase
Serum glutamic oxaloacetic transaminase
Serum glutamic pyruvic transaminase
Total protein
Albumin
Globulin
Albumin/Globulin ratio
Total cholesterol
Direct Bilirubin
Total Bilirubin
Calcium
Potassium
Lactic Dehydrogenase
Sodium
Chloride

Urinanalysis included:

Color	Protein	Occult Blood
Appearance	Glucose	Nitrite
pH	Ketones	Urobilinogen
Specific gravity	Bilirubin	

Gross Necropsy:

10 rats/sex/dose were killed at 12 and 18 months. The surviving animals were killed at 24 months. Thorough external and internal examinations were performed on all animals. The following tissues were weighed at scheduled sacrifices: adrenals, brain including stem, ovaries, testes, heart, kidneys, liver, pituitary, spleen, and the thyroid/parathyroid complex.

HISTOPATHOLOGY

The following tissues from all animals were sectioned, stained with hematoxylin and eosin, and examined microscopically:

abdominal aorta	heart (with coronary vessels)
adrenal (both)	intestine
bone and bone marrow	cecum
bone marrow smear	colon
blood smear	duodenum
brain	ileum
eyes with contiguous Harderian gland and optic nerve	jejunum (distal)
esophagus	skin, flank
	spinal cord (cervical and thoracic

5

kidneys (2)
liver (2 sections)
lung
lymph nodes
mammary gland
pancreas
paranasal passage
pituitary
prostate
major salivary glands

sciatic nerve with muscle
seminal vesicles (right)
skeletal muscle (biceps femoris)
spleen
stomach
thymus
trachea
thyroid/parathroid (complex)
urinary bladder
uterus (corpus and cervix uteri)
vagina
gross lesions
tissue masses or suspect tumors and regional lymph nodes.
ovaries (both
testes with epidiymides (both)

Pathology Personnel

Dr. R. J. Arceo, M.D., I.R.D.C. staff pathologist examined tissues from the 12 and 18 month interim sacrifices, from the rats that died in the first 18 months, and sections of duodenum from the terminal sacrifice. Tissues from the terminal sacrifice, and animals that died after 18 months were examined by Dr. L. W. Nelson, D.V.M., Ph.D., a subcontractor in Mount Vernon, Indiana. He also reexamined the duodenal sections from the final sacrifice.

RESULTS

Feed Analysis:

Concentrations of captan were analyzed in the control (none found) and test diets and generally were within 10% of the expected levels with the exception of the analysis at week 104 in which captan was found in decreased amounts (up to 18% lower at 25 mg/kg/day, and 14% lower at 250 mg/kg/day).

Food Consumption:

A slight dose-related decrease in food consumption was seen.

Dose (mg/kg/day)	Food Consumption (percent different from control) Males	Females
0	-	-
25	+1.6	-3.2
100	-2.4	-3.7
250	-5.1	-4.8

Mortality:

In male rats the survival was similar for controls and treated groups until approximately week 70 when the high dose group's survival relative to controls declined slightly. The mid dose group's survival dropped slightly at week 86 and the low dose group's survival decreased at week 98.

	Male Rats % survival at dose level (mg/kg/day)			
Week of Study	0	25	100	250
60	91	95	93	93
70	90	93	93	88
74	88	93	92	82
78	88	92	90	78
82	86	86	82	74
86	80	84	76	67
90	80	78	70	58
94	78	68	66	53
98	70	60	62	51
102	60	52	44	47
104	56	48	40	47

A generally similar pattern was observed with the females. A drop in survival compared to the controls was seen in the higher dose groups.

	Female Rats % survival at dose level (mg/kg/day)			
Week of Study	0	25	100	250
60	95	97	98	91
70	95	90	95	87
74	95	90	95	83
78	95	87	93	83
82	94	82	90	78
86	88	72	84	76
90	84	68	84	76
94	82	60	74	72
98	72	56	68	70
102	62	56	62	66
104	56	52	58	62

Body Weights:

There was a dose-related statistically significant decrease in mean body weights in both the mid and high dose groups throughout the study. The mean body weights in the low dose group were similar to the control values.

Week of Study	Mean body weights, % difference from control Males, dose levels				Females, dose levels			
	0	25	100	250	0	25	100	250
0	0	0	-3.0	-1.0	0	+1.1	+1.1	-1.1
26	0	-3.4	-7.1	-12.6	0	-1.6	-7.8	-9.4
52	0	-0.8	-5.7	-12.4	0	-4.2	-11.8	-17.3
78	0	-0.3	-8.5	-16.2	0	-6.6	-15.4	-18.9
104	0	+1.6	-11.9	-20.1	0	-6.5	-18.9	-19.4

General observations:

No trends indicating a dose related effect were reported. Localized hair loss, ulcerated body areas, and anogenital abdominal or thoracic masses were among the more frequently reported observations.

Ophthalmoscopic observations:

The summary stated that no obvious trends in pathology suggestive of test material-related reactions were observed. The report, however, did show a dose-related increase in chorioretinal degeneration and/or hyperplasia in male rats. In some cases, this condition spontaneously resolved during the study.

Localized Chorioretinal Degeneration and/or Chorioretinal Hyperplasia

	Dose (mg/kg/day) 0	25	100	250
Males				
Cases/group size	2/70	4/70	5/70	13/70
Cases that resolved	1	0	1	6
Cases seen at sacrifice	1	4	4	7
Females				
Cases/group size	3/70	6/70	3/70	3/70
Cases that resolved	1	2	1	2
Cases seen at sacrifice	2	4	1	2

Hematology:

No consistent change in the hematology values was seen that could be related to dose or duration of dosing. A few values differed statistically from the control values but no pattern was evident.

Blood biochemistry:

Generally no dose-related effect was seen. There was a statistically significant dose-related decrease in LDH at 12 months; however, this effect was not seen at other times. Increased BUN levels were reported at 24 months but no dose relationship is apparent. Statistically significant differences in Direct Bilirubin levels were reported but no trend is evident. Direct Bilirubin was not measured at 18 and 24 months "due to technical error."

Urinalysis:

No dose-related effect was seen in pH or specific gravity.

Gross necropsies:

No toxicologically significant macroscopic lesions were observed at gross necropsies.

Organ weights:

At the 12 month interim sacrifice, several statistically significant organ weight and organ weight relative to body weight increases were reported in males. Female rats showed statistically significant increases for relative organ weights at the high dose groups only for adrenals, kidneys (both), and heart.

Organ Weights at 12 Month Interim Sacrifice

Males

	Dose (mg/kg/day)			
	0	25	100	250
Average body weight (g)	755	686	653	607
Adrenals weight (mg)	51	69*	58	65
± S.D.	18.5	10.3	13.4	10.0
% relative weight	0.67	1.01*	0.90*	1.08*
Kidney, left (g)	2.43	2.60	2.72	2.69
% relative weight	0.32	0.38*	0.42*	0.45*
Kidney, right (g)	2.47	2.62	2.70	2.79
% relative weight	0.33	0.38	0.42*	0.47*
Heart (g)	1.72	1.85	1.76	1.69
% relative weight	0.23	0.27*	0.27*	0.28*

Brain (g)	2.23	2.33	2.23	2.16
% relative weight	0.30	0.34*	0.34*	0.36*

* Significantly different from the controls.

At the 18 month interim sacrifice statistically significant increases in absolute and relative weights were reported for livers at 250 mg/kg/day in males and females, kidneys in males at 250 mg/kg/day, and adrenals in males at 100 mg/kg/day. A statistically significant increase in relative heart weight was seen in males at 250 m/kg/day.

At the terminal sacrifice statistically significant changes in weights and relative weights were reported for the spleen, liver, kidneys, testes, heart, thyroid, and brain in both sexes.

Organ weights at final sacrifice

Dose (mg/kg/day)

	Males				Females			
	0	25	100	250	0	25	100	250
Average body weight (g)	640	648	563	504	430	405	341	345
Liver weight (g)	23.15	21.11	21.13	22.34	15.42	14.17	14.37	15.9
% relative weight	3.68	3.37	3.82	4.46*	3.62	3.48	4.23*	4.5
Kidney, left (g)	2.78	2.91	3.40*	3.21	1.70	1.56	1.70	1.7
% relative weight	0.46	0.48	0.62*	0.65*	0.41	0.39	0.52*	0.5
Kidney, right (g)	2.81	2.89	3.33	3.18	1.77	1.57*	1.75	1.7
% relative weight	0.46	0.47	0.60*	0.64*	0.42	0.40	0.53*	0.5
Heart (g)	2.16	2.15	2.12	2.11	1.61	1.51	1.43*	1.3
% relative weight	0.35	0.35	0.38	0.43*	0.38	0.38	0.43*	0.4
Thyroid/Parathyroid weight (mg)	47	58*	49	50	39	43	41	48
% relative weight	0.75	0.95	0.88	1.01*	0.93	1.10	1.23*	1.5
Brain weight (g)	2.25	2.30	2.21	2.15	2.06	2.03	1.99	2.0
% relative weight	0.37	0.37	0.40	0.49*	0.49	0.52	0.61*	0.6

* Significantly different from the controls.

Non-Neoplastic Histopathology:

The most notable effect seen was hepatocellular hypertrophy. A response was evident in males and females at 250 mg/kg/day at the eighteen month interim sacrifice and in males at 250 mg/kg/day (and possibly 100 mg/kg/day) at termination of the study when combining hypertrophy reported as "hepatocellular, centrolobular, and focal."

Chronic nephritis followed a dose response in the males early in the experiment up to eighteen months. All the males had chronic nephritis from the eighteen month interim sacrifice through the terminal sacrifice. Most of the females developed chronic nephritis by the end of the study, however no dose response was evident.

A dose response effect was seen for liver cytoplasmic vaculation in females that were sacrificed at 12 months, however no dose response was seen over the entire course of the study. Vaculation was reported in approximately half the rats by the end of the study.

A slight dose response for alveolar macrophage aggregates was suggested up to the eighteen months interim sacrifice, however by the end of the study, extensive lung histopathology indicative of chronic infection was reported for all dose groups equally.

No other non-neoplastic microscopic observations were increased over control incidences or showed a dose response trend.

Non-Neoplastic Histopathology

Dose (mg/kg/day)	12 month sacrifice + deaths		12-18 month deaths		18 month interim sacrifice		18 month to termination deaths		Total
	males	females	males	females	males	females	males	females	both
Hepatocellular Hypertrophy									
0	0/12*	0/12	0/5	0/1	0/10	0/10	2/43	7/47	9/140
25	0/13	0/11	0/2	0/8	0/10	0/10	2/45	5/41	7/140
100	0/13	0/11	0/3	0/3	0/10	0/10	8/44	8/46	16/140
250	0/14	0/14	1/9	2/6	10/10	8/10	12/38	5/39	38/140
Chronic Nephritis									
0	5/12	1/12	3/5	0/1	10/10	3/10	43/43	46/47	111/140
25	8/13	0/11	1/2	2/8	10/10	3/10	45/45	37/41	106/140
100	8/13	1/11	3/3	0/3	10/10	1/10	44/44	43/46	110/140
250	9/14	0/14	8/9	0/6	10/10	2/10	38/38	37/39	104/140
Liver Cytoplasmic Vaculation									
0	7/12	2/12	4/5	0/1	5/10	0/10	21/43	31/47	70/140
25	5/13	1/11	0/2	2/8	5/10	1/10	21/45	24/41	59/140
100	6/13	5/11	1/3	0/3	4/10	0/10	29/44	29/46	74/140
250	5/14	6/14	1/9	0/6	2/10	1/10	21/38	26/39	62/140
Alveolar Macrophage Aggregates									
0	3/12	4/12	1/5	0/1	9/10	5/10	—**	—	22/49**
25	1/13	2/11	0/2	3/8	4/10	4/10	—	—	14/54
100	5/13	3/11	2/3	2/3	7/10	2/10	—	—	21/50
250	4/14	6/14	4/9	4/6	7/10	4/10	—	—	29/63

* Number of responses/number of rats examined.

** Extensive focal lymphoid infiltrates, congestion, pneumonia, macrophage foci reported at all dose levels at 18 months to termination.

21

Histopathology: Neoplastic Lesions

No trends were evident for total numbers of tumors.

Male Rats With Tumors

	Dose (mg/kg/day)			
	0	25	100	250
Rats with tumors (%)	34(49%)	28(40%)	31(44%)	27(38%)
Average number of tumors per rat*	1.35	1.14	1.32	1.62
Rats with benign tumors (%)	29(41%)	25(36%)	26(37%)	24(34%)
Average number of benign tumors per rat*	1.31	1.12	1.34	1.25
Rats with malignant tumors (%)	8(11%)	4(6%)	6(9%)	4(6%)
Average number of malignant tumors per rat*	1.0	1.0	1.0	1.0

Female Rats With Tumors

	0	25	100	250
Rats with tumors (%)	48(69%)	43(61%)	50(71%)	50(71%)
Average number of tumors per rat*	1.90	1.58	1.98	1.68
Rats with benign tumors (%)	44(63%)	41(59%)	47(67%)	48(69%)
Average number of benign tumors per rat*	1.84	1.49	1.83	1.56
Rats with malignant tumors (%)	9(13%)	7(10%)	13(19%)	8(11%)
Average number of malignant tumors per rat*	1.0	1.0	1.0	1.13

* Average number of tumors in these rats having one or more tumors, rats without tumors are not included.

The most frequent tumor seen in both males and females was the adenoma (benign) in the pituitary.

Pituitary Adenomas

	Dose (mg/kg/day)			
	0	25	100	250
Total adenomas in male pituitaries (%)	16(23%)	13(19%)	18(26%)	15(21%)
Total adenomas in female pituitaries (%)	38(54%)	31(44%)	36(51%)	35(50%)

In contrast to studies in mice, no duodenal effects were seen.

A previous rat study (NCI, 1977) indicated that there may be significant trends associated with tumors in the adrenals and the thyroids. The following results were seen in this study:

Adrenal and Thyroid Tumors

	Dose (mg/kg) 0	25	100	250
Pheochromocytomas (benign) in male adrenals (%)	9(13%)	2(3%)	3(4%)	2(3%
Cortical cell tumors in female adrenals (%)	1(1%)[1]	0	1(1%)[2]	1(1%
Follicular cell adenomas in male thyroids (%)	2(3%)	0	0	4(6%
C-cell adenomas in male thyroids (%)	1(1%)	2(3%)	0	2(3%
Follicular cell adenomas in female thyroids (%)	0	0	0	1(1%
C-cell adenomas in female thyroids (%)	1(1%)	0	1(%)	2(3%

1. carcinoma
2. adenomas

A slight not statistically significant, increase in mammary gland tumors in females was reported.

Mammary Gland Tumors in Females

	0	25	100	250
intraductal papilloma	2	1	2	1
cystadenoma, papillary/adenoma	4	5	10	6
adenocarcinoma, papillary	1	3	3	5
Total: above tumors	7	9	15	12
Fibroadenoma	21	16	23	19

The only tumors for which a possible dose-effect relationship may be seen are kidney tumors in males.

Kidney Tumors in Males

	Dose (mg/kg/day)			
	0	25	100	250
Cortical cell/tubular adenoma in male kidneys (%)	1(1%)	0	2(3%)	3(4%)
Renal cell carcinoma/tubular adenocarcinoma in male kidneys (%)	0	1(1%)	1(1%)	1(1%)
Combined kidney tumors in males (%)	1(1%)	1(1%)	3(4%)	4(6%)

Discussion

This study was well performed in accordance with accepted protocols. Sufficient numbers of animals and dose groups were used. The study was well described with the exception of numerous pages that were marked as "revised" (signed by Edwin Goldenthal, Ph.D., Vice President and Director of Research, IRDC) dated 10/17/82, which was one year after the pathologist signed the report (L.W. Nelson, D.V.M., Ph.D., 7/29/81). These revised pages were found in the tumor tables, tissue inventory, materials and methods, and pathology reports. No explanation was given for these revisions or for the long time between completion of the study (terminal sacrifice, October 1-3, 1980) and its submission to EPA (received January 20, 1983).

Differences between the control and test animals do not appear to be related to the administration of captan except as described here:

1. Although the final survival at 104 weeks was similar in all control and dose groups for each sex, the higher dose groups tended to die sooner in a possible dose relationship.

2. There was a dose related decrease in mean body weights for both males and females in the mid and high dose levels throughout most of the study.

3. An increase (dose related) in chorioretinal degeneration and/or hyperplasia was seen in male rats by ophthalmoscopy but no effect was reported in the microscopic histopathology of sacrificed animals. It would seen that either the visual observations were spurious or that the condition was not detectable microscopically. The effects might have been localized and not evident on the histopathology sections.

4. The relative increase in liver weight in both sexes at the high dose and in females in the mid dose might be attributed to the hepatocellular hypertrophy. The increased hepatocellular

hypertrophy was seen at the high dose for both sexes at the 18 month interim sacrifice and at the mid and high doses in males at final sacrifice.

5. The tentative NOEL may be established at 25 mg/kg/day and the LEL is 100 mg/kg/day. The effects seen at 100 mg/kg/day include hepatocellular hypertrophy, increased relative organ weights for kidneys in males and females and heart, brain, liver, and thyroid/parathyroid in males, and a decrease in the mean body weights of males and females.

6. The kidney adenomas and carcinomas in the males need to be further evaluated.

William R. Schneider, Ph.D.
Toxicology Branch
Hazard Evaluation Division (TS-769)

TOX:SCHNEIDER:DCR-26151:pad:04/15/83:TOX-16
REVISED:05/5/83:DCR-17831
REVISED-07/12/83:DCR-11739:efs:TOX-16
REVISED-7/15/83:DCR-11280:efs:TOX-16

TOX:SCHNEIDER:DCR-26151:pad:04/15/83:TOX-16
REVISED:05/5/83:DCR-17831
REVISED-07/12/83:DCR-11739:efs:TOX-16