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

007802

MAR - 9 1990

MEMORANDUM

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

SUBJECT: Tribufos (DEF) Review of Teratology Studies in Rat and Rabbit and an Oncogenicity Study in mice.

TO: Jay Ellenberger PM-50
Registration Division (H7505C)

FROM: Robert P. Zendzian Ph.D. 2/23/90
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Compound; DEF | Tox Chem #864 | MRID #401906-01 & 02 411710-01

Registration #285465 285310 | Registrant; Mobay | Tox Project #0-0551 0-0328

Action Requested

Review the following studies;

Oncogenicity study of technical tribufos (DEF®) with mice. R.H. Hayes, Mobay, Corp Toxicology Depart. Study No. 86-27101, Report No. 99175, Jun 29, 1989, MRID 411710-01.

A teratogy study with DEF technical in the rat. R.L. Kowalski, Miles Laboratories Inc. Laboratory Report No. 87320, Aug 8, 1986, MRID 401906-01

A teratogy study with DEF technical in the rabbit. G.R. Clemens, J.J. Bare and R.E. Hartnagel Jr. Miles Laboratories Inc. Laboratory Report No. MTD0003, #94468, Jan 22, 1987, MRID 401906-02

Conclusions

MRID 411710-01. Guideline

The studies reviewed under this memorandum are the only acceptable toxicology studies on tribufos available to the Agency.

In an agency sponsured study, Abou-Donia (1978) determined that tribufos produced organophosphate type delayed neurotoxicity in hens after a single oral dose and with 90-day oral administration. Based on this report, tribufos was placed in RPAR (Rebutable Presumption Against Reregistration) status. Abou-Donia and coworkers subsiquently reported on the effects of route on tribufos neurotoxicity (1979A) and described a late acute toxic effect mediated by N-butyl mercaptan a metabolite of tribufos (1979b). This late acute toxicity is of particular importance as adiministration of atropine, which is recommended as antidotal to the toxicity of organophosphates, was lethal to the experimental animals during this late toxic effect.

During this RPAR process, Toxicology Branch indicated that the toxicology data base on tirbufos was deficient and recommended that certian studies be performed (Zendzian 1980). In November 1981 Special Review Division issued a decision document removing tribufos from the RPAR process and transfering it back to Registration Division. Zendzian (1981) commented that he could not defend this position since it lacked a requirement for performing the recommended toxicology.

On January 21, 1985 the Data Call-In Program issued a data call-in notice on tribufos requiring an oncogenicity study in the mouse (due 2/98) and teratology studies in two species (due 4/87). This notification was taken without informing Registration Division and Toxicology Branch.

In 1987 Registration Division informed Toxicology Branch, HED that no studies had been requested from the Registrant on DEF and requested a list of toxicology data requirements. Toxicology replied that no acceptable data was available on tribufos and, considering the existance of food tolerances the following studies were required (Zendzian 1987).

81-1 Acute Oral
81-2 Acute Dermal
81-3 Acute Inhalation
81-4 Primary Eye Irritation
81-5 Primary Dermal Irritation
81-6 Dermal Sensitization
81-7 Acute Delayed Neurotoxicity

82-1 Subchronic Oral, two species rodent and nonrodent
82-2 Subchronic Dermal (21-day)
82-4 Subchronic Inhalation
82-5 Subchronic Neurotoxicity

83-1 Chronic Toxicity, two species rodent and nonrodent
83-2 Oncogenicity, two species
83-3 Teratogenicity, two species
83-4 Reproduction

84-2 Mutagenicity Tests.

85-1 Metabolism

The memo also noted "-DEF shows significant differences in route-related toxicity. In addition to the oral doses specified in the guidelines, the metabolism of a single low dose dermal application on five male and five female rats should be determined. This is particularly important as the dermal route is the main route of exposure in man."

A hand written note on my copy of this memo indicated that the Agency had received teratology studies on tribufos in April 1987. However, these studies were not fowarded to Toxicology for review.

In March 1988 the Registrant held meetings with the Agency on the doses to be used in a chronic dog study, a rat reproduction study and a rat chronic/ oncogenicity study. These studies had been required by the State of California. Subsiquently the Registrant submitted protocols for the dog and rat chronic studies which were reviewed for acceptability (Zendzian 1988).

On February 3, 1989 the Registrant wrote a 6(a)(2) letter to the Agency on the results of a mouse oncogenicity study with tribufos. Statistically significant incidence of adenocarcinoma/carcinoma in the small intestine in both sexes, hemangiosarcoma in the liver of the males and alveolar/brochiolar neoplasia in the lungs of the females, all at the high dose, were observed (Zendzian 1989). The report of the study was submitted to the Agency in July 1989.

In December 1989, as part of FIFRA 88, Ester Saito of HED discovered the existance of the mouse oncogenicity study and the two teratology studies. Subsiquently they were sent to HED for review and are the subject of this memo.

Discussion

The rat and rabbit teratology studies are are acceptable and negative for fetal toxicity at the highest dose tested.

At the high dose, the mouse oncogenicity study showed a statistically significant incidence of adenocarcinoma/carcinoma in the small intestine in both sexes, hemangiosarcoma in the liver of the males and alveolar/brochiolar neoplasia in the lungs of the females. The information on tribufos will be presented to the HED Peer Review Committee for an evaluation and clasification of the oncogenicity of the compound.

Compound related effects were observed at all doses in both sexes. Effects are presented by dose below at the lowest dose at which they were observed.

Nominal dose of 10 ppm

Statistically significant decreases in plasma cholineterase activity at weeks 53, 78 and 90 all doses both sexes.

Statistically significant decreases in RBC cholineterase activity at weeks 78 and 90 all doses both sexes.

Statistically significant decreases in brain cholineterase activity at week 93 (termination) all doses in males.

At week 78 in the males, a significant decrease in mean cell volume and mean cell hemoglobin was observed at 10 and 50 ppm and significant decreases in red cell count, hemoglobin, hematocrit, mean cell volume and mean cell hemoglobin at 250 ppm. At week 90 in the males, a significant decrease in mean cell hemoglobin was observed at 50 ppm and significant decreases in red cell count, hemoglobin, hematocrit, mean cell volume and mean cell hemoglobin at 250 ppm. At week 90 in the females, a significant decrease in hematocrite was observed at 10 ppm and significant decreases in red cell count, hemoglobin and hematocrit at 50 and 250 ppm.

Nominal dose of 50 ppm

Statistically significant increased number of males showing paleness and hunched back.

Statistically significant histopathological observations are culled from the incidence table. [*$p \leq 0.05$]

	MALES Dose(ppm)				FEMALES Dose(ppm)			
Organ & Disease	Cont	10	50	250	Cont	10	50	250
Adrenals	50	50	50	50	50	50	50	49
Amyloid	5/50 3.8	6/50 3.3	15/50* 3.5	9/50 3.1	4/50 2.5	7/50 2.4	3/50 2.7	3/49 2.7
Epididymis	50	50	50	50				
Hyperspermatognensis, NOS	2/50 2.0	6/50 2,2	10/50* 2.1	3/50 2.0				
Small Intestine	50	50	50	50	50	50	50	50
Amyloid, NOS	6/50 2.7	7/50 2.6	20/50* 2.8	9/50 2.4	6/50 2.7	9/50 2.9	10/50 2.9	8/50 2.4
Degeneration, Vacuolar Epithelium		1/50 2.0	8/50* 2.1	28/50* 2.5			11/50 1.7	28/50 2.6
Spleen	50	50	50	50	50	50	49	50
Hematopoiesis, NOS	6/50 2.0	6/50 2.7	14/50* 2.8	19/50* 2.5	16/50 2.6	14/50 2.4	18/50 2.7	20/50 2.2

Nominal dose of 250 ppm

Statistically significant increased number of females showing loose stool and enlarged abdomen and males showing enlarged abdomen.

Increased mortality and concurrent decrease in life span in both sexes.

Increased mean food consumption and body weight in both sexes during the latter portion of the study.

Statistically significant histopathological observations are culled from the incidence table. Note particuarly the increased incidence of tumors in the liver, lung and small intestine. [* $p \leq 0.05$]

	MALES Dose(ppm)				FEMALES Dose(ppm)			
Organ & Disease	Cont	10	50	250	Cont	10	50	250
Adrenals	50	50	50	50	50	50	50	49
Calcification				4/50 1.0		2/50 1.5		5/49* 1.4
Degeneration/ pigmentation, NOS	17/50 1.4	15/50 1.6	21/50 2.0	39/50* 2.6	18/50 2.2	26/50 2.5	22/50 2.3	38/49* 2.7
Degeneration, NOS			1/50 1.0	22/50* 2.4	2/50 2.0		1/50 2.0	3/49 2.0
Caecum	50	50	50	50	50	50	50	50
Edema, NOS	4/50 2.0	6/50 2.0	5/50 2.4	10/50 2.1	6/50 2.0	3/50 2.0	4/50 1.8	17/50* 2.1
Liver	50	50	50	50	50	50	50	50
Hemangiosarcoma	1/50 M	1/50 M	4/50 M	7/50* M	2/50 M	2/50 M	2/50 M	1/50 M
Hypertropy, NOS	1/50 2.0		1/50 2.0	4/50 1.5		2/50 2.0		6/50* 1.8
Lung	50	50	50	50	50	50	50	50
Alveolar/bronchiolar adenoma	11/50 B	9/50 B	5/50 B	9/50 B	5/50 B	5/50 B	2/50 B	15/50* B
Mesenteric Lymph	48	50	48	46	49	50	50	50
Congestion, NOS	7/48 1.9	7/50 2.0	14/48 1.9	10/46 1.8	15/49 1.9	20/50 2.0	18/50 2.1	29/50* 1.9
Rectum	45	49	47	46	48	46	47	49
Inflammation, Acute NOS	3/45 1/7	2/49 2.0	3/47 2.0	11/46* 2.6	3/48 2.3		1/47 3.0	15/49 2.5
Necorsis, NOS		1/49 2.0		7/46* 2.6	2/48 2.5			7/49 2.9

Ulcer, NOS		1/49 2.0	1/47 3.0	10/46* 2.6	1/48 3.0		1/47 3.0	14/49* 2.9
Small Intestine	50	50	50	50	50	50	50	50
Adenocarcinoma, NOS				9/50* M		1/50 M		4/50 M
Dilated/Distended			2/50 3.0	7/50* 2.1	2/50 2.0		11/50* 2.0	28/50* 2.0
Hyperplasia, mucosa			1/50 2.0	22/50* 2.0	1/50 4.0			19/50* 2.3

Citations

Memo Zendzian to Brown SRP Division re Def and Merphos, Studies to be Requested from the Sponsers, Aug 12, 1980

Memo Zendzian to Hitch HED, re Decision Document, DEF dated November 1981, Nov 13, 1981

Memo Zendzian to Sanders FHB, RD re Merphos/DEF, Data Requirements July 20, 1987

Memo Zendzian to Taylor PM-25 RD, re DEF, Review of Protocol for Chronic Toxicity Study in Dogs and Chronic Toxicity/Oncogenicity Study in Rats, 9/9/88

Memo Zendzian to Taylor PM-25 RD, re DEF, (6)(a)(2) Submission, Positive Oncogenic Response in Mouse Oncogenicity Study 3/29/89

Attachments
DERs

cc
Taylor, PM-25 FHB, RD

Data Evaluation Report

007802

Compound Tribufos (DEF)

Citation

Oncogenicity study of technical tribufos (DEF®) with mice. R.H. Hayes, Mobay, Corp Toxicology Depart. Study No. 86-271-01, Report No. 99175, Jun 29, 1989, MRID 411710-01.

Reviewed by Robert P. Zendzian Ph.D. 1/30/90
Senior Pharmacologist

Core Classification Guideline

Conclusions

Mice dosed at 0, 10, 50 or 250 pmm for 90 weeks. At 10 ppm, decreased plasma and RBC cholinesterase both sexes, decreased brain cholinesterase males, at 78 weeks males decreased MCV and MCH, at week 90 females decreased hematocrite. At 50 ppm, males increased number showing paleness and hunched backs, at 78 weeks males decreased MCV and MCH, at week 90 decreased MCH, at week 90 females decreased RBC count, hemoglobin and hematocrite. Histopathology males; adrenals amyloid, epididymis hyperspermatogenensis, small intestine amyloid and vacuolar degeneration epithelium, spleen hematopoiesis. At 250 ppm loose stools females, enlarged abdomen both sexes, increased mortality/decreased life span both sexes, increased food consumption and body weight both sexes, decreased RCB count, hemoglobin, hematocrite, MCV and MCH in males, decreased RCB count, hemoglobin and hematocrite in females. Histopathology males, adrenals degeneration, liver hemangiosarcoma*, rectum acute inflammation, necrosis and ulcer, small intestine adenocarcinoma*, dilated/ distended and mucosal hyperplasis. In females, adrenals calcification and degeneration/pigmentation, caecum edema, liver hypertropy, lung alveolar/bronchiolar adenoma*, mesenteric lymph node congestion, rectum acute inflamation, necrosis and ulcer, small intestine adenocarcinoma*, dilated/distended, mucosal hyperplasia.

Materials

Technical grade tribufos (DEF)
S,S,S-Tributyl phosphorotrithioate
clear colorless liquid,
Batch No. 85-R-26-39
98.9% active
CAS 78-48-8

CD-1 mice (Crl:CD-1(ICR)BR) for Charles river

Experimental design

Fifty mice per sex per dose were dosed at nominal concentrations of 0, 10, 50 or 250 ppm tribufos in the diet for 90 weeks.

"All diets, incuding the control diet, were prepared using corn oil as the vehicle at one percent of the diet by weight."

A sample from each batch of diet and vehicle was analyzed for contaminents and the water was analyzed quarterly.

Test substance was analyzed prior to and periodically during the study.

Homogenecity and stability in the diet was determined and the concentration in the diet verified quarterly.

"Dose levels of 10, 50 and 2500 ppm were selected based on the results of an eight week pilot study on tribufos with mice. In the pilot study, the cholinesterase activity for mice consuming 270 ppm technical grade tribufos was inhibited 95 and 97% in plasma, 73 and 69& in erythrocytes and 26 and 29% in brain of males and females, respectively."

"Differental leucocyte counts were conducted on all survivers at 12 months, 18 months and at termination of the study or when animals were sacrificed in extremis.""-complete blood counts were performed on ten mice/sex/level at 12 and 18 months and at termination."

"Determinations for cholinesterase activity in plasma (PCHE) and erythrocytes (RCHE) were done at 12 and 18 months and at termination on ten mice/sex/dose. Brain cholinesterasse (BCHE) activity was determned for the same mice at termination of the study."

Gross pathology was performed on all mice found dead, sacrificed in extremis and at termination. The following tissues were collected for histopathological examination. Asterixed organs were weighed.

adrenals*
aorta, dorsal
bone (femur and vertebrae)
bone marrow
brain*
  cerebral cortex
  cerebellar cortex
  medulla/pons
cervex
clitoral gland
epididymis
esophagus
ovaries
pancreas
parathyroid
pituitary
preputial gland
prostate gland
ribs/costochondral junction
salivary glands, submaxillary
seminal vesicles
skin
skull-nasal cavity
small intestine

eyes
gall bladder
harderian gland
heart*
joint (femoral-tibial)
kidneys*
large intestine
  caecum
  colon
  rectum
larnyx
liver*
lungs*
lymph nodes
  cervical
  mysenteric
mammary gland
nerves
  optic
  sciatic
  duodenum
  jejunum
  ileum
spinal cord
  cervical
  thoracic
  lumbar
spleen*
sternum
stomach
testicles*
thymus
thyroid
trachea (with main stem bronchi)
urinary bladder
uterus
all gross lesions with a border of normal tisue

Results

Mean concentration of test compound in the diet was;

Nominal (ppm)	Actual (ppm)
10	8.6
50	44.9
250	233.0

Compound related observations, as number of mice showing the particular sign, consisted of;

	Males Nominal Dose (ppm)				Females Nominal Dose (ppm)			
	0	10	50	250	0	10	50	250
Loose stool	1/50	0/50	0/50	5/50	0/50	0/50	0/50	16/50*
Paleness	1/50	3/50	12/50*	13/50*	2/50	7/50	3/50	9/50
Hunch back	0/50	0/50	7/50*	4/50	6/50	6/50	7/50	11/50
Enlarged abdomen	4/50	5/50	9/50	17/50*	3/50	5/50	7/50	19/50*

* $p \leq 0.05$

Mortality was significantly increased in both sexes at 250 ppm (figures 1 and 2 from the report). Mean group survival times in days were as follows;

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Figure 1

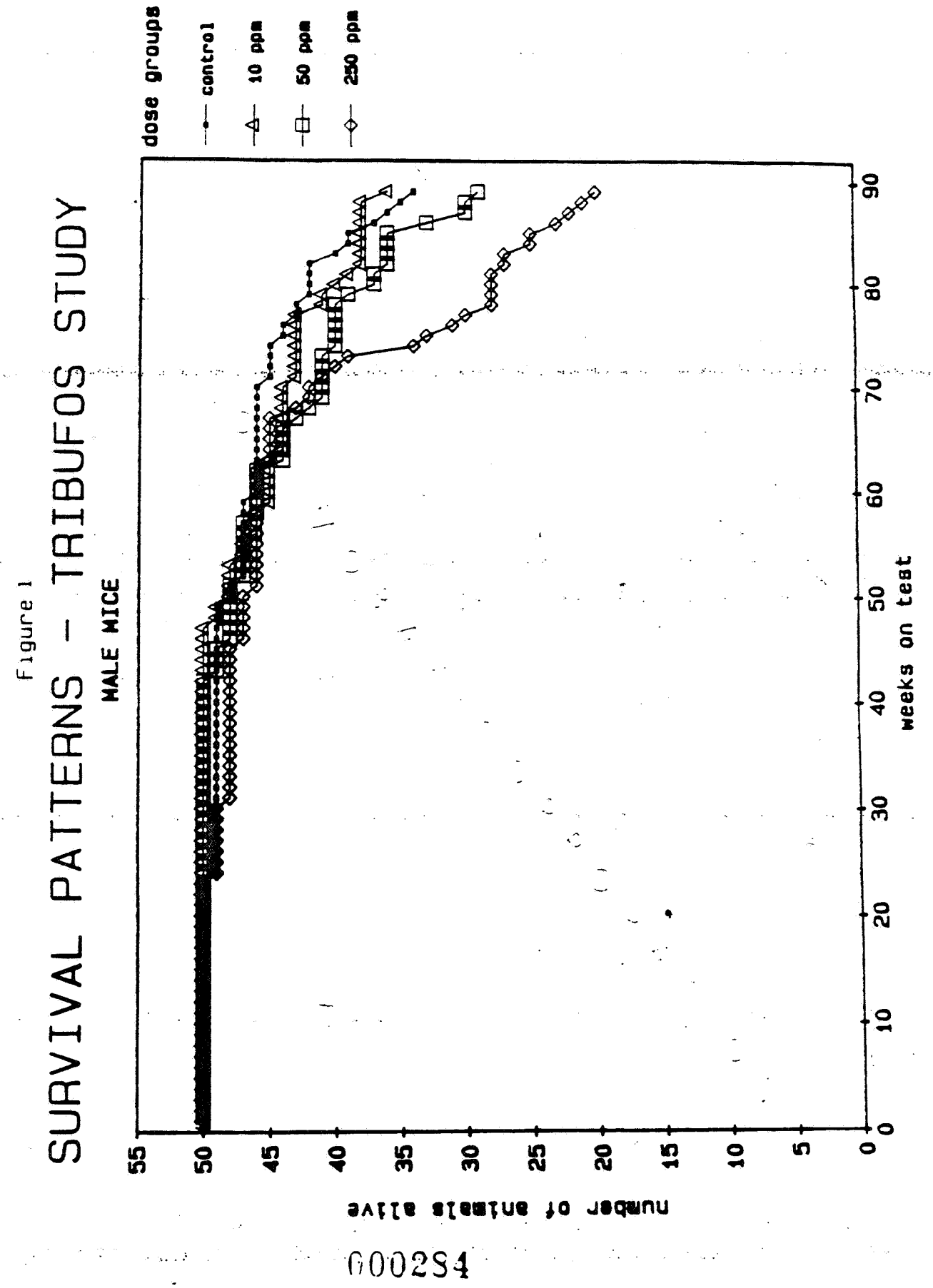

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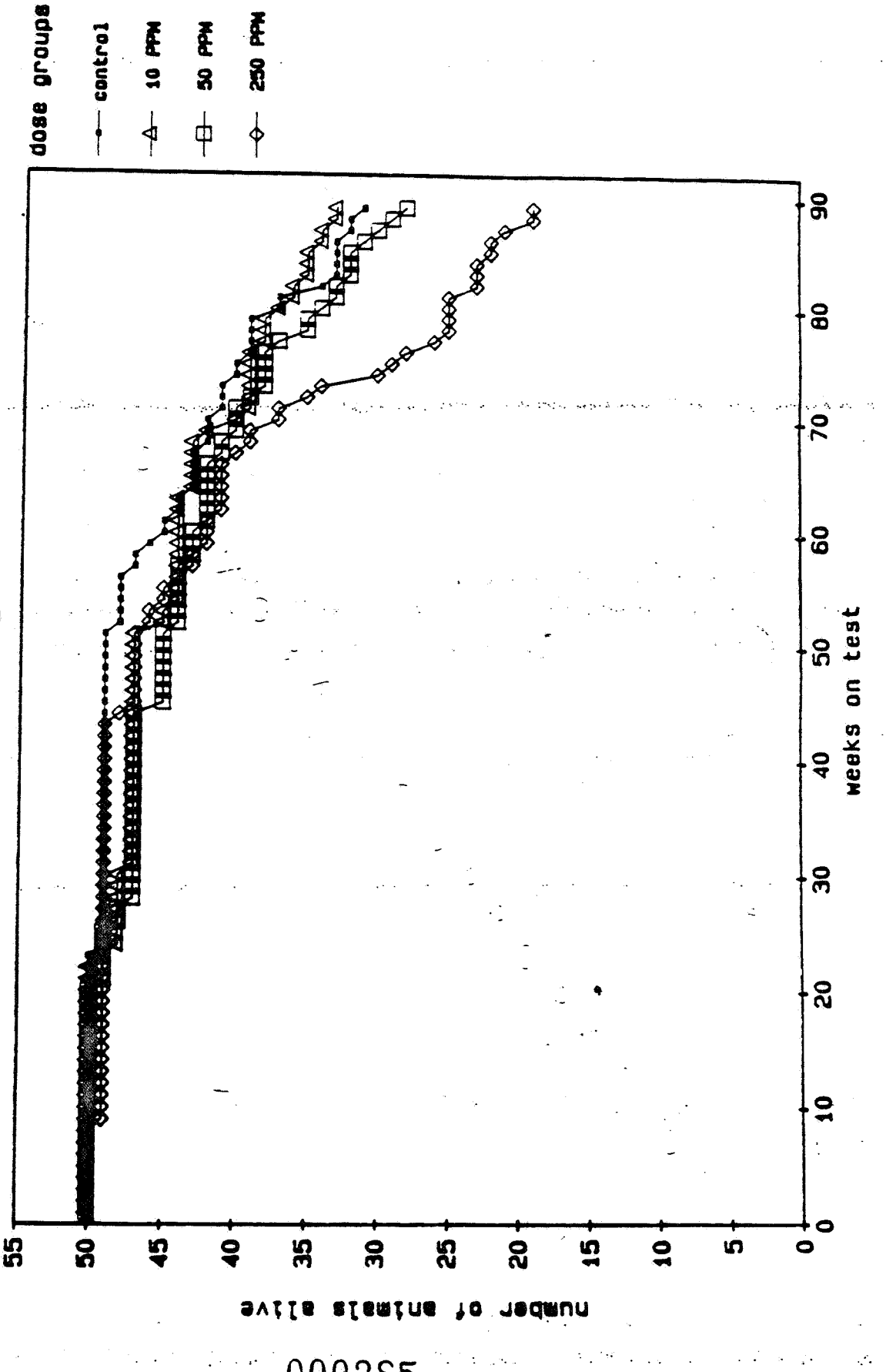
Figure 2

SURVIVAL PATTERNS - TRIBUFOS STUDY

FEMALE MICE



	Nominal Dose (ppm)			
	0	10	50	250
Males	595	592	580	553
Female	579	568	555	536

Mean food consumption was increased in both sexes at the high dose during the latter portion of the study (figures 3 and 4 from the report). Mean weekly body weights for the high dose group were significantly increased from week 13 to the end of the study (figures 5 and 6 from the report).

Mean daily intake of test compound, in mg/kg, was as follows;

	Nominal Dose (ppm)		
	10	50	250
Males	1.64	8.28	48.02
Female	2.08	11.14	63.04

Mean hematology values are presented in Table HE01, Hematology Summary Tables, from the report. At week 53 a significant decrease in hemoglobin was observed in the high dose females. At week 78 in the males, a significant decrease in mean cell volume and mean cell hemoglobin was observed at 10 and 50 ppm and significant decreases in red cell count, hemoglobin, hematocrit, mean cell volume and mean cell hemoglobin at 250 ppm. At week ninety in the males, a significant decrease in mean cell hemoglobin was observed at 50 ppm and significant decreases in red cell count, hemoglobin, hematocrit, mean cell volume and mean cell hemoglobin at 250 ppm. In the females, a significant decrease in hematocrite was observed at 10 ppm and significant decreases in red cell count, hemoglobin and hematocrit at 50 and 250 ppm.

Blood cholinesterase activity was as follows;

	Males		Females	
WEEK 53	PlasmaChe	RBCChe	PlasmaChe	RBCChe
control	4.35	1.61	6.36	1.47
10ppm	1.77*	1.59	2.67*	1.54
50ppm	0.54*	1.07*	0.35*	0.94*
250ppm	0.23*	0.75*	0.23*	0.86*

	Males		Females	
WEEK 78	PlasmaChe	RBCChe	PlasmaChe	RBCChe
control	4.56	1.62	4.86	1.60
10ppm	1.87*	1.51*	2.31*	1.43*
50ppm	0.48*	1.06*	0.47*	1.00*
250ppm	0.25*	0.74*	0.23*	0.76*

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Figure 3

FEED CONSUMPTION - TRIBUFOS

MALE MICE

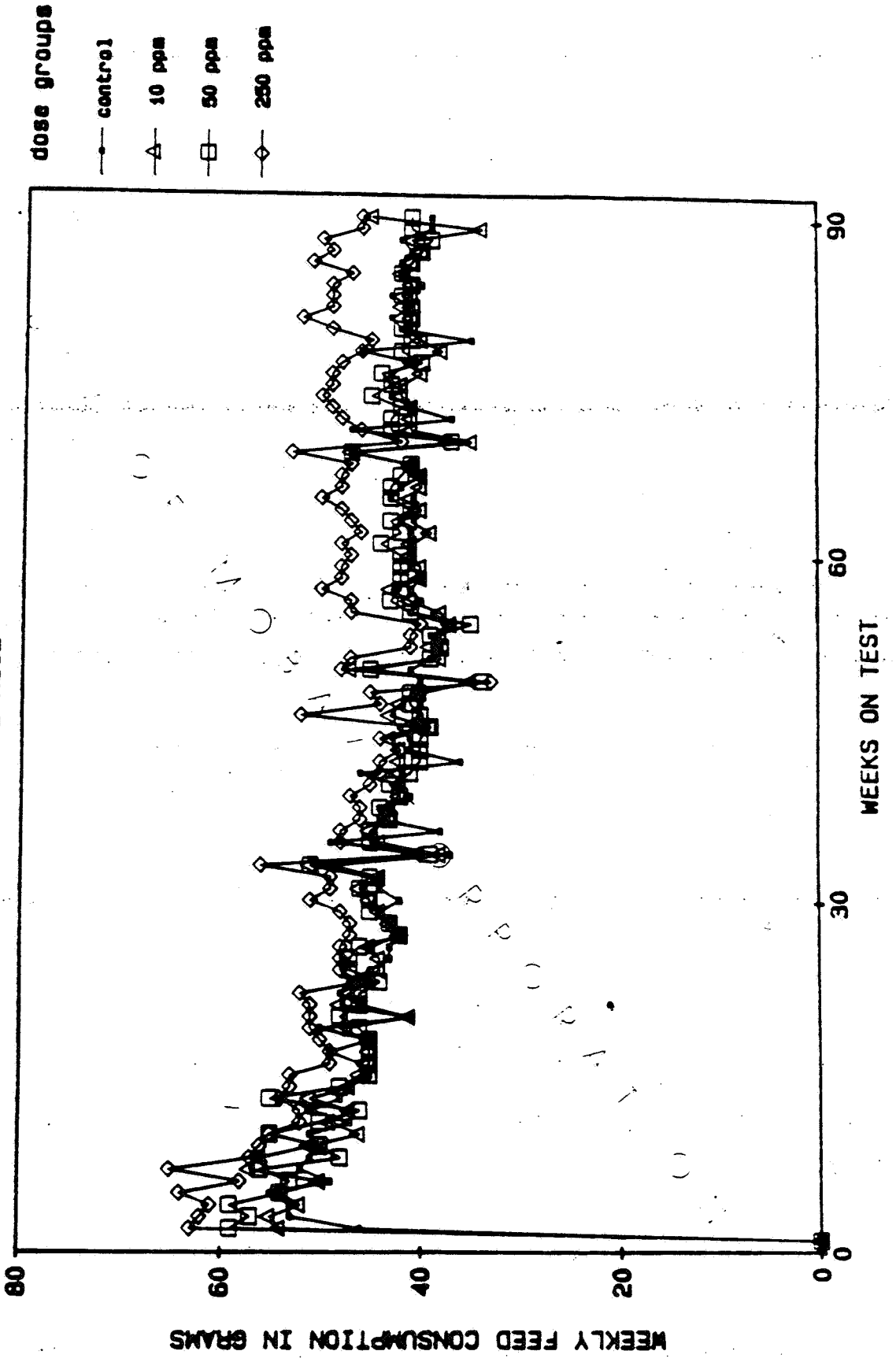

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Figure 4

FEED CONSUMPTION - TRIBUFOS

FEMALE MICE

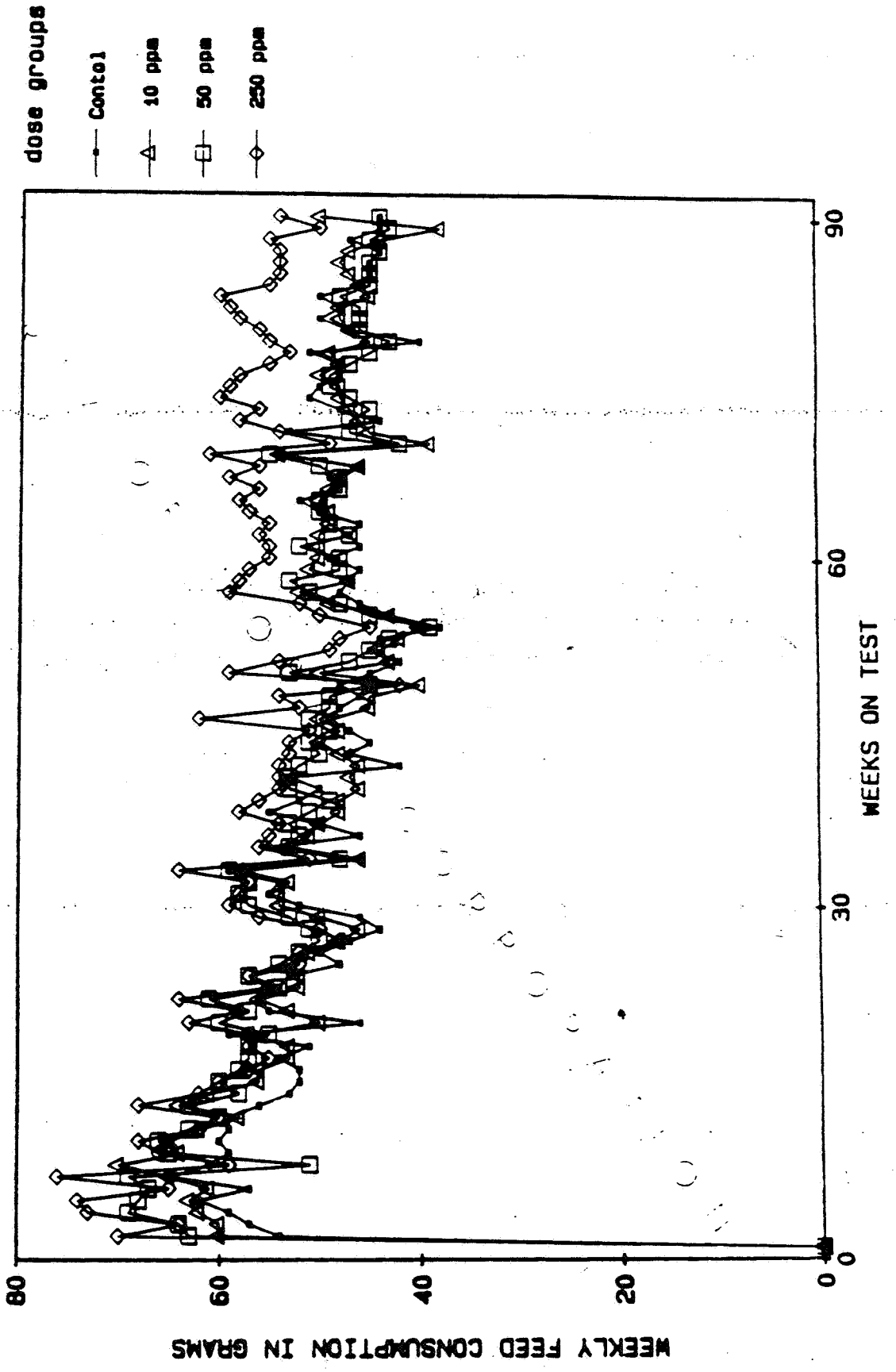

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Figure 5

BODY WEIGHTS - TRIBUFOS

MALE MICE

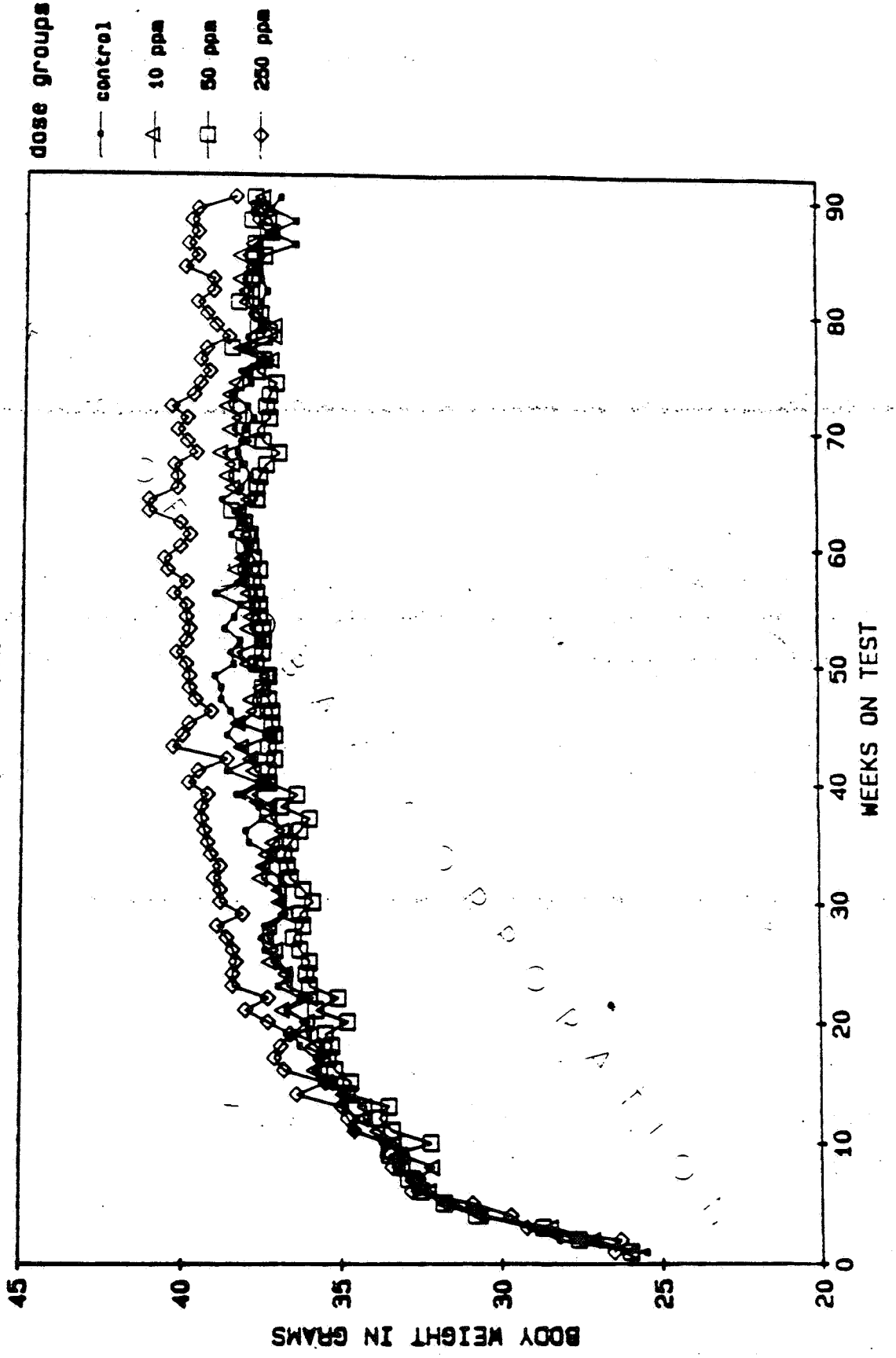

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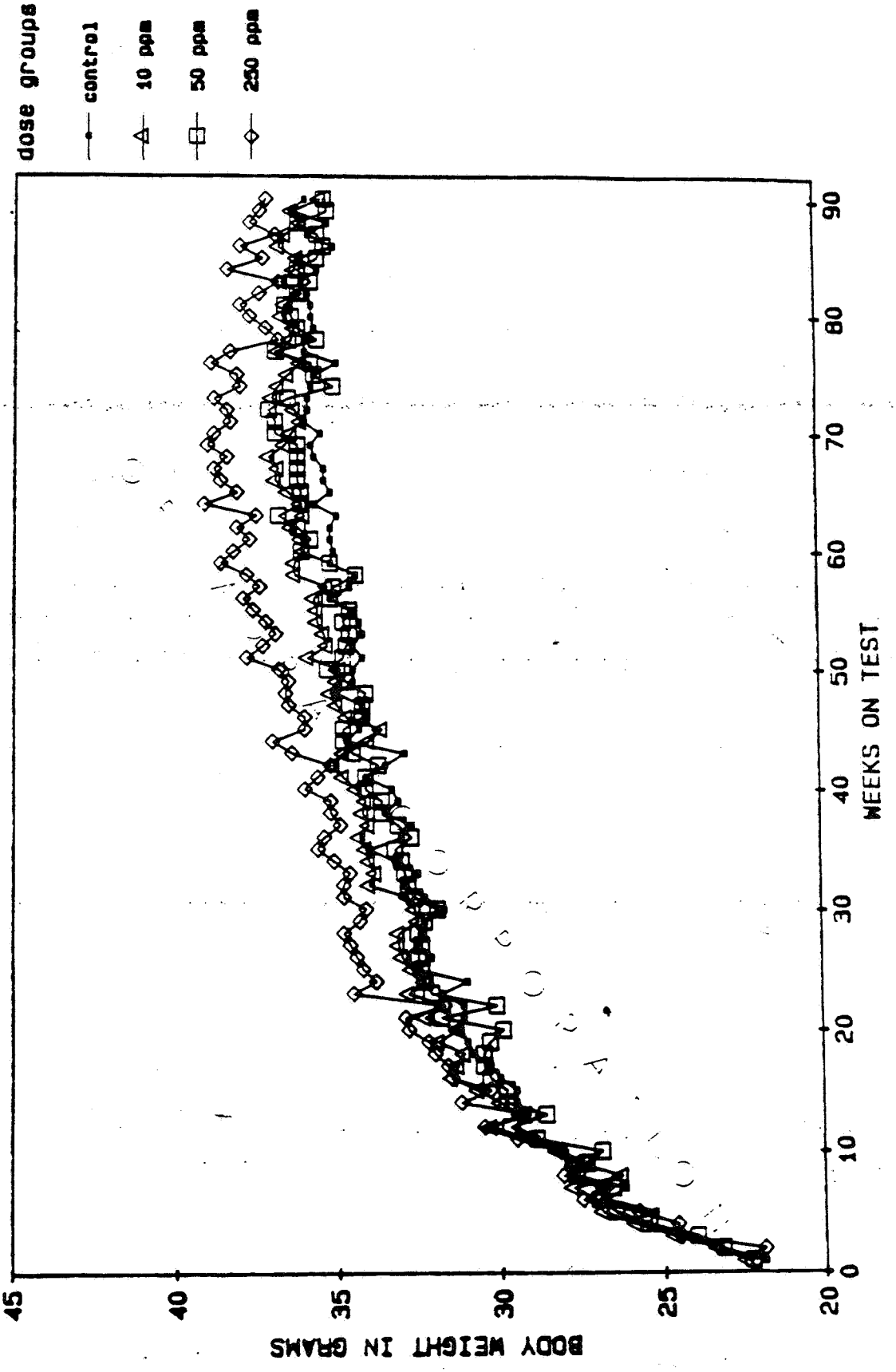
Figure 6

BODY WEIGHTS - TRIBUFOS

FEMALE MICE



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CORPORATE TOXICOLOGY DEPARTMENT

ABBREVIATIONS & DEFINITIONS FOR HEMATOLOGY

HEMATOLOGY PARAMETERS
- WBC = WHITE BLOOD CELLS (THOUSANDS PER CUBIC MM)
- RBC = RED BLOOD CELLS (MILLIONS PER CUBIC MM)
- HGB = HEMOGLOBIN (G/DL)
- HCT = HEMATOCRIT (PERCENT)
- MCV = MEAN CORPUSCULAR VOLUME (CUBIC MICRONS)
- MCH = MEAN CORPUSCULAR HEMOGLOBIN (PG)
- MCHC = MEAN CORPUSCULAR HEMOGLOBIN CONCENTRATION (G/DL)
- PLT = PLATELETS (THOUSANDS PER CUBIC MM)
- RETC = RETICULOCYTES (% OF RED BLOOD CELLS)

LEUKOCYTE DIFFERENTIAL COUNTS
- SEG = SEGMENTED GRANULOCYTE OR POLYMORPHONUCLEAR LEUKOCYTE
- BAND = BAND LEUKOCYTE
- LYMP = LYMPHOCYTE
- MONO = MONOCYTE
- EOSN = EOSINOPHIL
- BASO = BASOPHIL
- ATYP = ATYPICAL LYMPHOCYTE
- IMGR = IMMATURE GRANULOCYTE
- BLAS = BLAST CELL
- OTHR = OTHER CELLS
- NUCR = NUCLEATED RED BLOOD CELLS PER 100 WHITE BLOOD CELLS

ERYTHROCYTE MORPHOLOGY

MORPHOLOGY SYMBOLS: 1 = SLIGHT
2 = MODERATE
3 = MARKED

- POLC = POLYCHROMASIA
- HYPO = HYPOCHROMASIA
- POIK = POIKILOCYTOSIS
- TARG = TARGET CELLS
- SPHR = SPHEROCYTOSIS
- ANIS = ANISOCYTOSIS
- MICR = MICROCYTOSIS
- MACR = MACROCYTOSIS
- SIKL = SICKLE CELLS
- BSTP = BASOPHILIC STIPPLING
- VAC = VACUOLES
- TOXG = TOXIC GRANULATION

MOBAY CORPORATION
CORPORATE TOXICOLOGY DEPARTMENT

TABLE: HE01 - HEMATOLOGY SUMMARY TABLES
COMPOUND: DEF STUDY 88-271-01 SPECIES: MOUSE

HEMATOLOGY ANALYSES - WEEK 53

MEAN VALUES FOR MALES

LEVEL	WBC	RBC	HGB	HCT	MCV	MCH	MCHC	PLT
CONTROL	6.4	8.75	13.2	40.4	46.1	14.6	31.6	1054
10 PPM	5.2	8.94	14.1	42.1	47.1	15.6	33.1	1182
50 PPM	5.5	8.63	14.0	41.0	47.6	16.3	34.2	1268
250 PPM	7.6	8.13	12.7	37.9	46.7	15.7	33.6	1508*

LEVEL	SEG	BAND	LYMP	MONO	EOSN	BASO	ATYP	IMGR	BLAS	OTHR	NUCR	RETC
CONTROL	27	1	65	6	1	0	1	0	0	0	0	-
10	27	1	65	6	1	0	1	0	0	0	0	-
50	30	1	64	5	0*	0	0	0	0	0	0	-
250	23	0	71*	4*	0*	0	1	0	0	0	0	-

MEAN VALUES FOR FEMALES

LEVEL	WBC	RBC	HGB	HCT	MCV	MCH	MCHC	PLT
CONTROL	4.8	8.91	14.6	41.8	47.1	16.4	34.9	1103
10 PPM	6.2	9.30	15.0	42.4	46.6	16.1	34.7	1293
50 PPM	5.1	8.65	14.3	41.1	47.6	16.5	34.7	1329
250 PPM	5.8	8.84	13.6*	42.1	47.7	15.4*	32.3*	1333

LEVEL	SEG	BAND	LYMP	MONO	EOSN	BASO	ATYP	IMGR	BLAS	OTHR	NUCR	RETC
CONTROL	25	1	69	4	0	0	1	0	0	0	0	-
10 PPM	22	0	72	5	0	0	1	0	0	0	0	-
50 PPM	22	1	71	5	1*	0	1	0	0	0	0	-
250 PPM	22	1	73	3	0	0	0	0	0	0	0	-

* = SIGNIFICANTLY (P<0.05) DIFFERENT THAN CONTROL VALUE

MOBAY CORPORATION
CORPORATE TOXICOLOGY DEPARTMENT

COMPOUND: DEF

TABLE: HE01 - HEMATOLOGY SUMMARY TABLES
STUDY 86-271-01

SPECIES: MOUSE

HEMATOLOGY ANALYSES - WEEK 78

MEAN VALUES FOR MALES

LEVEL	WBC	RBC	HGB	HCT	MCV	MCH	MCHC	PLT
CONTROL	9.6	9.27	13.7	41.3	44.5	14.9	33.4	1714
10 PPM	8.9	8.82	13.8	40.8	46.4*	15.7*	33.9	1551
50 PPM	8.7	8.40	13.5	39.5	47.2*	16.1*	34.1	1517
250 PPM	8.4	7.32*	11.7*	34.9*	47.9*	16.0*	33.6	1439

LEVEL	SEG	BAND	LYMP	MONO	EOSN	BASO	ATYP	IMGR	BLAS	OTHR	NUCR	RETC
CONTROL	24	1	66	6	1	0	2	0	0	0	0	-
10	31	0	60	5	0	0	2	0	0	0	0	-
50	30	1	62	6	0*	0	2	0	0	0	0	-
250	26	0	69	3*	0*	0	2	0	0	0	0	-

MEAN VALUES FOR FEMALES

LEVEL	WBC	RBC	HGB	HCT	MCV	MCH	MCHC	PLT
CONTROL	4.7	8.26	13.2	39.3	48.0	16.1	33.6	1078
10 PPM	6.5	8.80	14.0	41.0	46.6	16.0	34.3*	1341
50 PPM	5.3	8.10	13.1	38.1	47.1	16.2	34.4*	1208
250 PPM	5.8	7.71	12.4	37.0	48.8	16.3	33.5	1249

LEVEL	SEG	BAND	LYMP	MONO	EOSN	BASO	ATYP	IMGR	BLAS	OTHR	NUCR	RETC
CONTROL	28	0	66	5	0	0	2	0	0	0	0	-
10 PPM	28	0	62	6	1	0	3	0	0	0	0	-
50 PPM	22	0	71	4	0	0	2	0	0	0	0	-
250 PPM	26	0	69	3*	1	0	1	0	0	0	0	-

* = SIGNIFICANTLY (P<0.05) DIFFERENT THAN CONTROL VALUE

MOBAY CORPORATION
CORPORATE TOXICOLOGY DEPARTMENT

TABLE: HE01 - HEMATOLOGY SUMMARY TABLES
COMPOUND: DEF STUDY 88-271-01 SPECIES: MOUSE

HEMATOLOGY ANALYSES - WEEK 90

MEAN VALUES FOR MALES

LEVEL	WBC	RBC	HGB	HCT	MCV	MCH	MCHC	PLT
CONTROL	6.6	9.04	12.9	40.0	44.3	14.0	31.6	1394
10 PPM	6.9	8.82	13.6	40.5	46.0	15.5	33.7	1441
50 PPM	6.3	8.02	12.7	37.5	47.1	15.8*	33.7	1445
250 PPM	7.3	6.46*	10.6*	32.2*	51.2*	16.8*	32.8	1798

LEVEL	SEG	BAND	LYMP	MONO	EOSN	BASO	ATYP	IMGR	BLAS	OTHR	NUCR	RETC
CONTROL	24	1	68	5	1	0	1	0	0	0	0	-
10	28	1	66	5	0	0	2	0	0	0	0	-
50	24	1	68	5	0	0	2	0	0	0	0	-
250	27	1	68	4	0	0	1	0	0	0	0	-

MEAN VALUES FOR FEMALES

LEVEL	WBC	RBC	HGB	HCT	MCV	MCH	MCHC	PLT
CONTROL	5.5	9.08	14.2	41.9	46.3	15.7	33.8	1343
10 PPM	6.1	8.60	13.5	39.0*	45.4	15.7	34.7*	1281
50 PPM	4.7	8.22*	13.1*	38.4*	47.0	16.0	34.1	1134*
250 PPM	6.2	7.88*	12.3*	37.4*	47.8	15.7	32.9*	1336

LEVEL	SEG	BAND	LYMP	MONO	EOSN	BASO	ATYP	IMGR	BLAS	OTHR	NUCR	RETC
CONTROL	26	1	65	5	0	0	2	0	0	0	0	-
10 PPM	23	1	67	7	0	0	2	0	0	0	0	-
50 PPM	20	1	72	4	1	0	2	0	0	0	0	-
250 PPM	18	5	72	3	1	0	1	0	0	0	0	-

* = SIGNIFICANTLY ($P \leq 0.05$) DIFFERENT THAN CONTROL VALUE

WEEK 90	Males		Females	
	PlasmaChe	RBCChe	PlasmaChe	RBCChe
control	5.07	1.64	6.70	1.50
10ppm	1.68*	1.35*	2.36*	1.23*
50ppm	0.48*	0.95*	0.46*	0.95*
250ppm	0.31*	0.74*	0.19*	0.75*

* P<0.05

Terminal (week 93) brain cholinesterase activity was as follows;

	Males	Females
control	17.1	15.9
10ppm	15.6*	15.3
50ppm	14.9*	15.5
250ppm	10.8*	11.6*

* P<0.05

External observations, during gross pathology, of enlarged abdomen and pale color were apparently treatment related. (see below). Abnormalities and masses observed during necropsy showed no relationship with treatment.

	MALES Dose(ppm)				FEMALES Dose(ppm)			
External Observation	Cont	10	50	250	Cont	10	50	250
Enlarged abdominal	4/50	3/50	11/50	12/50	5/50	6/50	7/50	13/50
Pale	2/50	4/50	9/50	13/50	6/50	8/50	8/50	14/50

observed/# examined

Selected histopathology incidences are summarized. Specific pathology was selected on the basis of dose-relation, statistical significance and/or relationship to pathological observations in the organ/tissue.

	MALES Dose(ppm)				FEMALES Dose(ppm)			
Organ & Disease	Cont	10	50	250	Cont	10	50	250
Adrenals	50	50	50	50	50	50	50	49
Amyloid	5/50 3.8	6/50 3.3	15/50* 3.5	9/50 3.1	4/50 2.5	7/50 2.4	3/50 2.7	3/49 2.7
Calcification				4/50 1.0		2/50 1.5		5/49* 1.4
Degeneration/ pigmentation, NOS	17/50 1.4	15/50 1.6	21/50 2.0	39/50* 2.6	18/50 2.2	26/50 2.5	22/50 2.3	38/49* 2.7
Degeneration, NOS			1/50 1.0	22/50* 2.4	2/50 2.0		1/50 2.0	3/49 2.0

observed/# examined, mean of severity codes(1-5), Benign, Malignant Not Otherwise Specified, *<0.05

Histopathology summary (cont)

Organ & Disease	MALES Dose(ppm) Cont	10	50	250	FEMALES Dose(ppm) Cont	10	50	250
Caecum	50	50	50	50	50	50	50	50
Edema, NOS	4/50 2.0	6/50 2.0	5/50 2.4	10/50 2.1	6/50 2.0	3/50 2.0	4/50 1.8	17/50* 2.1
Epididymis	50	50	50	50				
Hyperspermatognensis, NOS	2/50 2.0	6/50 2,2	10/50* 2.1	3/50 2.0				
Eyes	50	50	50	50	50	50	50	50
Calcification, NOS	8/50 1.8	8/50 1.9	9/50 1.9	4/50 1.3	7/50 1.9	8/50 1.4	13/50 1.8	12/50 1.9
Liver	50	50	50	50	50	50	50	50
Hemangiosarcoma	1/50 M	1/50 M	4/50 M	7/50* M	2/50 M	2/50 M	2/50 M	1/50 M
Hepatocelular Adenoma	4/50 B	3/50 B	2/50 B		1/50 B	1/50 B		
Hepatocelular Carcinoma, NOS	7/50 M	4/50 M	2/50 M	2/50 M		1/50 M	1/50 M	1/50 M
Hyperplasia, Focal		3/50 3.0					1/50 3.0	
Hypertropy, NOS	1/50 2.0		1/50 2.0	4/50 1.5		2/50 2.0		6/50* 1.8
Inflamation, chronic active	13/50 1.9	14/50 2.0	9/50 2.3	2/50 2.0	8/50 2.1	7/50 2.1	9/50 2.3	
Inflamation, acute NOS	2/50 2.5	5/50 2.4	5/50 3.0	3/50 2.0	14/50 2.0	2/50 2.5	6/50 1.5	3/50 2.0
Lung	50	50	50	50	50	50	50	50
Alveolar/bronchiolar adenoma	11/50 B	9/50 B	5/50 B	9/50 B	5/50 B	5/50 B	2/50 B	15/50* B
Hyperplasia, focal	4/50 3.0	2/50 3.0	2/50 3.0	6/50 2.5	3/50 2.7	4/50 3.0	3/50 2.7	8/50 2.8
Hyperplasia, Lymphoid NOS	8/50 2.1	9/50 2.0	7/50 1.9	4/50 1.5	17/50 1.9	10/50 2.1	18/50 2.1	11/50 2.0

observed/# examined, mean of severity codes(1-5), Benign, Malignant
Not Otherwise Specified, *<0.05

Histopathology summary (cont)

Organ & Disease	MALES Dose(ppm) Cont	10	50	250	FEMALES Dose(ppm) Cont	10	50	250
Mesenteric Lymph	48	50	48	46	49	50	50	50
Congestion, NOS	7/48 1.9	7/50 2.0	14/48 1.9	10/46 1.8	15/49 1.9	20/50 2.0	18/50 2.1	29/50* 1.9
Hyperplasia, Lymphoid NOS	5/48 2.0	6/50 2.3	5/48 2.0	9/46 2.4	6/49 2.7	8/50 2.5	7/50 2.4	4/50 2.3
Multiple Organs	2	4	5	9	11	7	10	5
Malignant Lymphoma		2/4 M	3/5 M	4/9 M	8/11 M	5/7 M	7/10 M	5/5 M
Rectum	45	49	47	46	48	46	47	49
Inflammation, Acute NOS	3/45 1/7	2/49 2.0	3/47 2.0	11/46* 2.6	3/48 2.3		1/47 3.0	15/49 2.5
Necorsis, NOS		1/49 2.0		7/46* 2.6	2/48 2.5			7/49 2.9
Ulcer, NOS		1/49 2.0	1/47 3.0	10/46* 2.6	1/48 3.0		1/47 3.0	14/49* 2.9
Small Intestine	50	50	50	50	50	50	50	50
Adenocarcinoma, NOS				9/50* M		1/50 M		4/50 M
Amyloid, NOS	6/50 2.7	7/50 2.6	20/50* 2.8	9/50 2.4	6/50 2.7	9/50 2.9	10/50 2.9	8/50 2.4
Autolysis	3/50 2.7	1/50 2.0	4/50 2.0	10/50 2.2	4/50 2.5			2/50 2.5
Degeneration, Vacuolar Epithelium		1/50 2.0	8/50* 2.1	28/50* 2.5			11/50 1.7	28/50 2.6
Dilated/Distended			2/50 3.0	7/50* 2.1	2/50 2.0		11/50* 2.0	28/50* 2.0
Hyperplasia, mucosa			1/50 2.0	22/50* 2.0	1/50 4.0			19/50* 2.3
Spleen	50	50	50	50	50	50	49	50
Hematopoiesis, NOS	6/50 2.0	6/50 2.7	14/50* 2.8	19/50* 2.5	16/50 2.6	14/50 2.4	18/50 2.7	20/50 2.2

observed/# examined, mean of severity codes(1-5), Benign, Malignant
Not Otherwise Specified, *<0.05

Discussion

Compound related effects were observed at all doses in both sexes. Effects are presented by dose below at the lowest dose at which they were observed.

Nominal dose of 10 ppm

Statistically significant decreases in plasma cholineterase activity at weeks 53, 78 and 90 all doses both sexes.

Statistically significant decreases in RBC cholineterase activity at weeks 78 and 90 all doses both sexes.

Statistically significant decreases in brain cholineterase activity at week 93 (termination) all doses in males.

At week 78 in the males, a significant decrease in mean cell volume and mean cell hemoglobin was observed at 10 and 50 ppm and significant decreases in red cell count, hemoglobin, hematocrit, mean cell volume and mean cell hemoglobin at 250 ppm. At week 90 in the males, a significant decrease in mean cell hemoglobin was observed at 50 ppm and significant decreases in red cell count, hemoglobin, hematocrit, mean cell volume and mean cell hemoglobin at 250 ppm. At week 90 in the females, a significant decrease in hematocrite was observed at 10 ppm and significant decreases in red cell count, hemoglobin and hematocrit at 50 and 250 ppm.

Nominal dose of 50 ppm

Statistically significant increased number of males showing paleness and hunched back.

Statistically significant histopathological observations are culled from the incidence table. [*$p \leq 0.05$]

	MALES Dose(ppm)				FEMALES Dose(ppm)			
Organ & Disease	Cont	10	50	250	Cont	10	50	250
Adrenals	50	50	50	50	50	50	50	49
Amyloid	5/50 3.8	6/50 3.3	15/50* 3.5	9/50 3.1	4/50 2.5	7/50 2.4	3/50 2.7	3/49 2.7
Epididymis	50	50	50	50				
Hyperspermatognensis, NOS	2/50 2.0	6/50 2,2	10/50* 2.1	3/50 2.0				

Small Intestine	50	50	50	50	50	50	50	50
Amyloid, NOS	6/50 2.7	7/50 2.6	20/50* 2.8	9/50 2.4	6/50 2.7	9/50 2.9	10/50 2.9	8/50 2.4
Degeneration, Vacuolar Epithelium		1/50 2.0	8/50* 2.1	28/50* 2.5			11/50 1.7	28/50 2.6
Spleen	50	50	50	50	50	50	49	50
Hematopoiesis, NOS	6/50 2.0	6/50 2.7	14/50* 2.8	19/50* 2.5	16/50 2.6	14/50 2.4	18/50 2.7	20/50 2.2

Nominal dose of 250 ppm

Statistically significant increased number of females showing loose stool and enlarged abdomen and males showing enlarged abdomen.

Increased mortality and concurrent decrease in life span in both sexes.

Increased mean food consumption and body weight in both sexes during the latter portion of the study.

Statistically significant histopathological observations are culled from the incidence table. Note particularly the increased incidence of tumors in the liver, lung and small intestine. [*$p \leq 0.05$]

	MALES Dose(ppm)				FEMALES Dose(ppm)			
Organ & Disease	Cont	10	50	250	Cont	10	50	250
Adrenals	50	50	50	50	50	50	50	49
Calcification				4/50 1.0		2/50 1.5		5/49* 1.4
Degeneration/ pigmentation, NOS	17/50 1.4	15/50 1.6	21/50 2.0	39/50* 2.6	18/50 2.2	26/50 2.5	22/50 2.3	38/49* 2.7
Degeneration, NOS			1/50 1.0	22/50* 2.4	2/50 2.0		1/50 2.0	3/49 2.0
Caecum	50	50	50	50	50	50	50	50
Edema, NOS	4/50 2.0	6/50 2.0	5/50 2.4	10/50 2.1	6/50 2.0	3/50 2.0	4/50 1.8	17/50* 2.1
Liver	50	50	50	50	50	50	50	50
Hemangiosarcoma	1/50 M	1/50 M	4/50 M	7/50* M	2/50 M	2/50 M	2/50 M	1/50 M

Hypertropy, NOS	1/50 2.0		1/50 2.0	4/50 1.5		2/50 2.0		6/50* 1.8
Lung	50	50	50	50	50	50	50	50
Alveolar/bronchiolar adenoma	11/50 B	9/50 B	5/50 B	9/50 B	5/50 B	5/50 B	2/50 B	15/50* B
Mesenteric Lymph	48	50	48	46	49	50	50	50
Congestion, NOS	7/48 1.9	7/50 2.0	14/48 1.9	10/46 1.8	15/49 1.9	20/50 2.0	18/50 2.1	29/50* 1.9
Rectum	45	49	47	46	48	46	47	49
Inflammation, Acute NOS	3/45 1/7	2/49 2.0	3/47 2.0	11/46* 2.6	3/48 2.3		1/47 3.0	15/49 2.5
Necorsis, NOS		1/49 2.0		7/46* 2.6	2/48 2.5			7/49 2.9
Ulcer, NOS		1/49	1/47	10/46*	1/48		1/47	14/49*
Small Intestine	50	50	50	50	50	50	50	50
Adenocarcinoma, NOS				9/50* M		1/50 M		4/50 M
Dilated/Distended			2/50 3.0	7/50* 2.1	2/50 2.0		11/50* 2.0	28/50* 2.0
Hyperplasia, mucosa			1/50 2.0	22/50* 2.0	1/50 4.0			19/50* 2.3

Data Evaluation Report

00/802

Compound Tribufos (DEF)

Citation

A teratogy study with DEF technical in the rat. R.L. Kowalski, Miles Laboratories Inc. Laboratory Report No. 87320, Aug 8, 1986, MRID 401906-01

5/20/90

Reviewed by Robert P. Zendzian Ph.D.
Senior Pharmacologist

Core Classification Guideline

Conclusions

Pregnant rats were dose orally at 0, 1, 7 and 28 mg/kg/day (days 6-16). Maternal RBC and plasma cholinesterase activity was depressed at 7 and 28 mg/kg/day and brain activity at 28 mg/kg/day. Maternal weight gain was decreased at 28 mg/kg/day. Maternal toxicity LEL 7 mg/kg/day, NOEL 1 mg/kg/day. Fetotoxic NOEL 28 mg/kg/day (HDT).

Materials

DEF technical
98% active
Batch No. 85-R-26-39 supplied by Mobay Corp.

Charles River Crl:CD®(SD)BR rats from Charles River

Experimental Design

Females were housed overnight with breeder males (2/male) and vaginal smears taken. The day of positive smear was designated day zero. Inseminated females were assigned randomly to the following test groups.

Dose mg/kg	Termination Phase I Day 16	Termination Phase II Day 20
vehicle	5	28
1	5	28
7	5	28
28	5	28

Test material was prepared as a 0.2% w/v emulsion in an aqueous CMV vehicle and administered orally on days 6 through 15 of gestation. Test animals were observed daily for signs of toxicity and weighed on days 0, 6, 8, 10, 12, 15 and 20 of gestation. Food consumption was recorded on days 1, 6, 8, 12, 15 and 20.

On day 16 phase I animals were weighed, sacrificed and blood and brain collected for determination of cholinesterase activity. "The abdominal and thoracic viscera from each dam were examined, pregnancy confirmed and any gross changes recorded."

"On day 20 of gestation, 5 days after the last dose of the test or control article, all phase II dams were weighed and sacrificed by CO_2 asphyxiation. Blood and brain tissue were collected from the first 10 dams found to be pregnant in each dose group, for measurement of erythrocyte, plasma and brain Che activity. The abdomen of each dam was opened, ovaries were excised and corpora lutea graviditatis counted and recorded. The intact uterus was transected at the cervix, trimmed along the antimesometrial margin, removed, and weighed. Each uterine horn was longitudionally opened and the fetuses displaced to one side to facilitate inspection of the uterine walls for the presence of implants and resorptions. After removing all fetuses and resorptions and recording each implant, the uteri were pressed between 2 glass plates to assure that all implantation scars had been noted. The abdominal and thoracic viscera from each dam were scrutanized and any gross changes were recorded."

"Fetuses were removed from their embryonic membranes, the umbilical cord was severed close to its attachment to the fetus and viability of the fetus was determined. Each fetus was blotted dry, removing blood and amniotic fluid, sexed and weighed. Individual placentas corresponding to each fetus were cleaned of extraneous tissue, blotted dry and weighed. A complete external examination was made of each fetus. ---------- Brains were obtained form the first 20 fetuses per dose group (the first fetuses selected for gross visual examination from 20 different litters), including the control group, for the purpose of measuring fetal brain Che activity.--------- Following removal of the brain a complete internal examination was conducted on the thoracic and abdominal viscera from these fetuses."

Aproximately one-half of the fetuses from each dam were examined internally. "Following visceral examination the fetuses were placed in Bouin's fixative and later free-hand razor blade sections were made through eyes and cranium." The remaining fetuses were prepared and examined for bone development and abnormalities.

Results

No treatment-related effects were observed clinically with the exception of two high dose animals observed salivating on study days 9 and 12. No effects were observed on food consumption. Mean weight gain during gestation was significantly reduced in the high dose group (Table III from the report).

Mean percent cholinesterase inhibition is summarized in Table V from the report. In the phase I dams (gestation day 16) statistically significant inhibition was observed in the RBCs and plasma at 7 and 28 mg/kg/day and in the brain at 28 mg/kg/day. In the phase II dams (gestation day 20) statistically significant inhibition was observed in the RBCs at 7 and 28 mg/kg/day and in the brain at 28 mg/kg/day. No effect on brain cholinesterase was observed in the phase II fetuses

Table VI from the report summarizes the dam reproductive efficiency and fetal data. A slight decrease in the fertility index was observed at the high dose (82.1% versus 92.9% controls) but it was not statistically significant. No other compound-related effects were observed.

Table VII from the report summarizes the distribution of resorptions in dams. No compound-related effects were observed.

Table VIII from the report summarizes the mean values of reproductive parameters of the females. No compound-related effects were observed.

Table IX from the report presents the external and viserial findings on the fetuses at termination. No compound-related effects were observed.

Table X from the report summarizes the skeletal variations/ abnomalities observed. No compound-related effects were observed.

Table III

Mean Body Weights (grams) of Pregnant Dams[a] During Gestation

	Control			1 mg/kg			7 mg/kg			28 mg/kg		
Day	Mean	S.E.	N	Mean	S.E.	N	Mean	S.E.	N	Mean	S.E.	N
0	259.2	4.8	26	264.6	4.2	27	265.9	3.9	27	262.6	5.0	23
6	282.0	5.3	26	284.9	4.2	27	287.7	3.9	27	284.1	5.3	23
8	287.6	5.4	26	293.7	4.5	27	296.4	4.1	27	291.0	5.6	23
10	296.4	5.5	26	303.1	4.7	27	306.0	4.2	27	287.0	5.5	23
12	306.7	5.5	26	312.6	4.9	27	314.7	4.5	27	297.7	5.7	23
15	323.2	5.7	26	330.7	5.2	27	332.2	5.0	27	311.4	6.5	23
20	390.4	7.6	26	401.0	6.4	27	403.2	6.1	27	373.9	9.9	23
Gain	131.2	4.9	26	136.4	4.2	27	137.3	3.7	27	111.3*	6.9	23.
%Gain	50.6			51.6			51.6			42.4		
Actual Wt.	311.1	5.9	26	318.0	4.9	27	320.3	5.0	27	293.3	6.1	23
Actual Gain	51.9	2.8	26	53.5	2.7	27	54.3	2.8	27	30.8**	2.3	23
%Gain	20.0			20.2			20.4			11.7		

[a] Includes only dams with viable fetuses on Day 20

* Significantly less than control at the .05 level using Dunnett's test

** Significantly less than control at the .01 level using Dunnett's test

Table V

Cholinesterase Inhibition (%)

Gestation Day	Dose mg/kg	N	Plasma	Erythrocyte	Dam Brain	Fetal Brain
16	0	5	0	0	0	-
16	1	5	27.6[a]	-17.3	6.4	-
16	7	5	57.6[a]	71.2[a]	17.2	-
16	28	5	74.7[a]	87.3[a]	57.6[a]	-
20	0	10	0	0	0	0
20	1	10	16.9	12.3	-7.0	0
20	7	10	12.5	39.6[a]	4.9	0
20	28	10	24.4	50.6[a]	46.4[a]	-2.7

[a]Significantly different from the control at the 0.05 level (Dunnett's test)

Table VI

Dam Reproductive Efficiency and Fetal Data
Phase II

		Control	1 mg/kg	7 mg/kg	28 mg/kg
No. of Pregnant Dams/Total		26/28	27/28	27/28	23/28
Fertility Index		92.9	96.4	96.4	82.1
Cestation Index		100	100	100	100
No. of Litters		26	27	27	23
No. with Resorption Sites Only		0	0	0	0
No. of Deaths Among Dams		0	0	0	0
No. of Dams Which Aborted		0	0	0	0
No. of Corpora Lutea	Median	17.0	17.0	17.0	17.0
	(Range)	(5-20)	(14-30)	(13-28)	(0-22)
Total No. of Implantations		384	431	429	343
	Median	16.0	17.0	17.0	16.0
	(Range)	(1-19)	(6-19)	(6-19)	(1-22)
Total No. of Fetuses		362	406	405	324
Litter Size	Median	15.5	15.0	16.0	15.0
	(Range)	(1-18)	(6-19)	(6-19)	(1-22)
Median Percent Male Fetuses		46.1	50.0	52.9	50.0
Median Wt. Viable Fetuses (gm)	(Male)	3.7	3.6	3.6	3.8
	(Female)	3.5	3.5	3.4	3.6
	(Combined)	3.6	3.5	3.5	3.7
Median Wt. of Placentas		0.52	0.54	0.53	0.54
No. of Resorption Sites	Median	1.0	1.0	1.0	1.0
	(Range)	(0-3)	(0-4)	(0-3)	(0-3)
Total No. of Dead Fetuses		0	0	0	0
	(Range)	(0-0)	(0-0)	(0-0)	(0-0)
% Pre-implantation Loss	Median	0	0	0	0
	(Range)	(0-80.0)	(0-66.7)	(0-78.6)	(0-56.3)
% Post-implantation Loss	Median	5.6	5.3	5.3	5.9
	(Range)	(0-23.1)	(0-25.0)	(0-27.3)	(0-17.6)

Table VII

Distribution of Resorptions in Dams

No. of Resorptions	Number of Dams with Resorptions			
	Control	1 mg/kg	7 mg/kg	28 mg/kg
0	11	13	12	11
1	9	7	8	6
2	5	4	5	5
3	1	2	2	1
4	0	1	0	0
Total No. of Resorptions	22	25	24	19
No. of Dams with More than 1 Resorption	6	7	7	6
Percent with Resorptions	57.7	51.9	55.6	52.2

Table VIII

Mean Values of Reproductive Parameters of Females

	Control	1 mg/kg	7 mg/kg	28 mg/k
No. of Corpora Lutea	16.1	17.2	17.3	15.2
(Range)	(5-20)	(14-30)	(13-28)	(0-2
Litter Size	13.9	15.0	15.0	14.
(Range)	(1-18)	(6-19)	(6-19)	(1-2
No. of Resorptions	0.8	0.9	0.9	0
(Range)	(0-3)	(0-4)	(0-3)	(0
No. of Implantations	14.8	16.0	15.9	1
(Range)	(1-19)	(6-19)	(6-19)	(1
% Pre-implantation Loss	12.3	8.4	7.5	
(Range)	(0-80.0)	(0-66.7)	(0-78.6)	(0
% Post-implantation Loss	5.7	5.8	5.8	
(Range)	(0-23.1)	(0-25.0)	(0-27.3)	((

Table IX

External and Visceral Findings on Fetuses at Termination (Day 20)

Dose[a]	Dam No.	Fetus No.	Observation
Control	RS4414	1170	Kidney, rt, pelvis dilated
	RS4466	1190	No innominate artery; rt subclavian and rt carotid arteries branch together directly from aorta
	RS4515	1201	No innominate artery; rt subclavian and rt carotid arteries branch separately from aorta
	RS4559	1210	Brain, ventricles, bilateral dilation
1	RS4423	570	Eye, lt, microphthalmia
	RS4428	347	Ureter, lt, distended
	RS4443	359	Ureter, lt, distended
	RS4450	1404,1406	Brain diminished in size
	RS4481	85	Brain, craniorhechischisis; vertebral column appears pinched together between thoracic and lumbar regions; amniotic sac filled with blood
		83	No innominate artery; rt subclavian and rt carotid arteries branch separately from aortic arch
	RS4503	616	Ureter, lt, distended
	RS4517	1054,1055	Placenta's, fused
7	RS4419	267	Kidney, pelves, bilateral dilation; ureters, bilateral distension
		274	Ureter, lt, distended
	RS4431	293	Kidney, pelves, bilateral dilation
	RS4457	117,120	Kidney, pelves, bilateral dilation; ureters, bilateral distension
		115	Ureter, lt, distended
	RS4475	1297	Brain, diminished in size
		1294	No innominate artery; rt subclavian and rt carotid arteries branch together from aorta
	RS4478	304	Brain. ventricle, bilateral dilation
	RS4516	877	Testis, rt, rudimentary
28	RS4445	240	Kidney, lt, pelvis dilated; ureter, lt, distended
	RS4452	907	Brain, ventricle, bilateral dilation
	RS4459	1479	Runt (1.9 g)
	RS4465	1349	Brain, ventricle, bilateral dilation
	RS4470	243	Posterior portion of fetus appears under-developed (posterior from lumbar region); did not show up skeletally; amniotic sac filled with blood

[a] mg/kg

Table X

Fetuses with One or More Skeletal Variations

Dose:	Control		1 mg/kg		7 mg/kg		28 mg/kg	
No. of Specimens Examined:	187		211		208		164	
Variation and/or Abnormality	No.	%	No.	%	No.	%	No.	%
SKULL: BONES INCOMPLETELY OSSIFIED	113	60.4	131	62.1	140	67.3	81	49.4
SKULL: SUTURES ENLARGED	20	10.7	26	12.3	17	8.2	12	7.3
SKULL: FONTANELLE ENLARGED	27	14.4	35	16.6	31	14.9	18	11.0
SKULL: PRESENCE OF CALCIFIED BODY			1	0.5	1	0.5	1	0.6
HYOID: VARIATIONS OF HYOID BODY	33	17.6	42	19.9	39	18.8	14*	8.5
RIBS: EXTRA RIBS	7	3.7	2	0.9	1	0.5	3	1.8
RIBS: INCOMPLETELY OSSIFIED			2	0.9			1	0.6
RIBS: WAVY OR CURVED			4	1.9	1	0.5	4	2.4
RIBS: RUDIMENTARY OR OC	6	3.2	2	0.9	3	1.4	4	2.4
RIBS: SMALL	13	7.0	12	5.7	7	3.4	11	6.7
VERTEBRAE: CERVICAL-ARCHES INCOMP. OSS,	10	5.3	5	2.4	23	11.1	8	4.9
VERTEBRAE: THORACIC-ARCHES FUSED			1	0.5				
VERTEBRAE: THORACIC-ARCHES MISSING			1	0.5				
VERTEBRAE: THORACIC-CENTRA INCOMP. OSS.	122	65.2	148	70.1	148	71.2	123	75.0
VERTEBRAE: THORACIC-CENTRA UNOSSIFIED							1	0.6
VERTEBRAE: THORACIC-CENTRA FUSED			1	0.5				
VERTEBRAE: THORACIC-CENTRA BIPARTITE	1	0.5	2	0.9	3	1.4	4	2.4
VERTEBRAE: THORACIC-CENTRA MISSING			1	0.5				
VERTEBRAE: LUMBAR-ARCHES FUSED			1	0.5				
VERTEBRAE: LUMBAR-ARCHES MISSING	2	1.1	2	0.9	2	1.0	6	3.7
VERTEBRAE: LUMBAR-CENTRA INCOMP. OSS.	3	1.6	8	3.8	3	1.4	6	3.7
VERTEBRAE: LUMBAR-CENTRA FUSED			1	0.5				
VERTEBRAE: LUMBAR-CENTRA MISSING	1	0.5	2	0.9	2	1.0	6	3.7
VERTEBRAE: SACRAL-ARCHES INCOMP. OSS.	104	55.6	154**	73.0	160**	76.9	92	56.1
VERTEBRAE: SACRAL-ARCHES UNOSSIFIED	5	2.7			2	1.0	3	1.8
VERTEBRAE: SACRAL-ARCHES SHIFT	2	1.1	3	1.4	2	1.0	4	2.4
VERTEBRAE: SACRAL-CENTRA INCOMP. OSS.	1	0.5			1	0.5		
VERTEBRAE: SACRAL-CENTRA UNOSSIFIED							1	0.6
VERTEBRAE: CAUDAL-ARCHES INCOMP. OSS.	24	12.8	23	10.9	30	14.4	9	5.5
VERTEBRAE: CAUDAL-ARCHES UNOSSIFIED	12	6.4	13	6.2	22	10.6	4	2.4
VERTEBRAE: CAUDAL-CENTRA UNOSSIFIED	1	0.5			1	0.5	1	0.6
PELVIS: ILIUM UNALIGNED	1	0.5	3	1.4	2	1.0	4	2.4
PELVIS: ISCHIUM INCOMPLETELEY OSSIFIED	3	1.6	2	0.9	1	0.5	3	1.8
PELVIS: ISCHIUM UNALIGNED	1	0.5	2	0.9	2	1.0	4	2.4
PELVIS: PUBIS UNOSSIFIED					1	0.5		
PELVIS: PUBIS INCOMPLETELY OSSIFIED	8	4.3	5	2.4	17	8.2	4	2.4

(continued)

Table X

Fetuses with One or More Skeletal Variations

Dose:	Control		1 mg/kg		7 mg/kg		28 mg/kg	
No. of Specimens Examined:	187		211		208		164	
Variation and/or Abnormality	No.	%	No.	%	No.	%	No.	%
STERNEBRAE: 1ST-INCOMPLETELY OSSIFIED	30	16.0	21	10.0	26	12.5	7**	4.3
STERNEBRAE: 2ND-UNOSSIFIED	3	1.6			4	1.9		
STERNEBRAE: 2ND-INCOMPLETELY OSSIFIED	64	34.2	71	33.8	86	41.3	32**	19.5
STERNEBRAE: 3RD-INCOMPLETELY OSSIFIED	38	20.3	29	13.8	34	16.3	16**	9.8
STERNEBRAE: 3RD-BIPARTITE					1	0.5		
STERNEBRAE: 4TH-UNOSSIFIED	1	0.5			4	1.9	1	0.6
STERNEBRAE: 4TH-INCOMPLETELY OSSIFIED	144	77.0	165	78.6	177	85.1	139	84.8
STERNEBRAE: 5TH-UNOSSIFIED	68	36.4	75	35.9	94	45.4	70	42.7
STERNEBRAE: 5TH-INCOMPLETELY OSSIFIED	119	63.6	132	63.2	111	53.6	93	56.7
STERNEBRAE: 5TH-BIPARTITE			1	0.5				
STERNEBRAE: 6TH-UNOSSIFIED	11	5.9	7	3.3	20	9.7	8	4.9
STERNEBRAE: 6TH-INCOMPLETELY OSSIFIED	139	74.3	178*	85.2	169	81.6	137	83.5
STERNEBRAE: 6TH-ASYMMETRICAL	1	0.5						
APPENDAGES: ANTERIOR-UO METACARPALS							1	0.6
APPENDAGES: ANTERIOR-IO METACARPALS	3	1.6			4	1.9	4	2.4
APPENDAGES: ANTERIOR-UO PHALANGES	6	3.2	5	2.4	10	4.8	3	1.8
APPENDAGES: POSTERIOR-UO METATARSALS	1	0.5	1	0.5	1	0.5	2	1.2
APPENDAGES: POSTERIOR-IO METATARSALS	2	1.1			2	1.0	2	1.2
APPENDAGES: POSTERIOR-UO PHALANGES	30	16.0	43	20.4	62**	29.8	22	13.4
SKULL: BONES ABNORMALLY SHAPED			1	0.5				
SKULL: BONES MISSING			1	0.5				
SKULL: BONES ABNORMALLY MALPOSITIONED			1	0.5				
SKULL: SUTURES MISSING			1	0.5				
VERTEBRAE: CERVICAL-ARCH ABN. POSITION			1	0.5				
VERTEBRAE: THORACIC-ARCH ABN. & MALPOS.			1	0.5				
VERTEBRAE: THORACIC-CENTRA ABN. & MAL.			1	0.5				
VERTEBRAE: LUMBAR-ARCH ABN. & MALPOS.			1	0.5				
VERTEBRAE: LUMBAR-CENTRA ABN. & MALPOS.			1	0.5				
VERTEBRAE: SACRAL-ARCH ABN. & MALPOS.			1	0.5				
VERTEBRAE: PINCHED FROM T 10 TO L3			1	0.5				
STERNEBRAE: 1ST-ABNORMALLY SHAPED			1	0.5				

* Significantly different than control at the 0.05 level
** Significantly different than control at the 0.01 level

Data Evaluation Report

Compound Tribufos (DEF) 007802

Citation

A teratogy study with DEF technical in the rabbit. G.R. Clemens, J.J. Bare and R.E. Hartnagel Jr. Miles Laboratories Inc. Laboratory Report No. MTD0003, #94468, Jan 22, 1987, MRID 401906-02

1/20/90

Reviewed by Robert P. Zendzian Ph.D.
Senior Pharmacologist

Core Classification Guideline

Conclusions

Pregnant rabbits were dosed at 0, 1, 3 or 9 mg/kg/day, days 7-19. Plasma and RBC cholinesterase activity was significantly reduced at all doses on day 20 and RBC at all doses on day 28. Does failed to gain weight at 9 mg/kg/day during dosing. Maternal toxicity LEL 9 mg/kg/day, NOEL 3 mg/kg/day. Fetal toxicity NOEL 9 mg/kg/day (HDT).

Materials

DEF technical
98% active
Batch No. 85-R-26-39 supplied by Mobay Corp.

American Dutch Rabbits from Langshaw farms

Experimental Design

Young adult female rabbits (> 4.5 months) weighing 2.32 to 3.51 kg were primed with HCG and artificially inseminated over a four day period. Does were ranbomly assigned to treatment groups of control, 1, 3, or 9 mg/kg/day, 17 does per group.

The test compound was prepared as a 0.022, 0.067 or 0.2% emulsion in an aqueous CMC (0.5% w/v carboxymethycelulose and 0.4% w/v polysorbate 80 in distiled water) vehicle. Solutions were analyzed for concentration and stability. Test material was administered in a constant volume of 4.5 ml/kg body weight.

Test material was administered orally on days 7 through 19 of gestation.

Does were observed daily for morbidity and morality.

Body weights were abtained on days 0, 7, 10, 14, 19, 21 and 28 of gestation. Food consumption was measured on days 1, 6, 8, 12, 15, 20, 23 and 28 of gestation.

On day 20 of gestation (24 hours after the last dose of test compound) and on day 28 blood was obtained for RBC and plasma cholinesterase determinations. At termination, on day 28, half the brain of each doe was collected for brain cholinesterase determination.

"On day twenty-eight of gestation, all does were sacrificed by intravenous barbiturate overdose. The abdomen was opened, ovaries were excised and corpora lutea were counted and recorded. The uterine horns were transected at the cervix, removed, and weighed. Each uterine horn was longitudinally opened along the antimesometrial surface and the amniotic sacs displaced to one side to facilitate inspection of the uterine walls for the presence of resorptions. All fetuses and resorptions were removed and each implant was noted. The abdominal and thoracic viscera from the does were scrutinized and gross anatomical changes were recorded."

Each fetuses was removed from its amniotic sac, the umbilical cord was severed close to its attachment to the fetus and viability of the fetus was determined. Placentas were removed, trimmed and weighed. Each fetus was blotted dry, removing blood and amniotic fluid and weighed. A complete external examination was made of each fetus. ------A complete internal examination was conducted on the thoracic and abdominal viscera and sex was determined for all fetuses." The head was skinned to view the eyes and a cross section made through the cerebrum.

All fetuses were prepared and examined for bone development and abnormalities.

Results

Clinical signs related to treatment were not observed. Two low dose does and one high dose doe died of resporatory disease. One low dose doe and one high dose doe aborted during the study.

Mean group body weights are summarized in Table 1 from the report. A statistically significant decrease in mean weight gain was observed in the high dose does during dosing. These does failed to gain weight during this period. No compound-related effects were observed on food consumption.

Results of cholinesterase determinations are summarized in Table III from the report. Plasma and RBC cholinesterase activity was significantly depressed at 20 days of gestation in all dose groups. At 28 days RBC cholinesterase activity remained significantly depressed at all doses. No compound-related effect was observed in the brain at termination.

Table IV from the report summarizes reproduction efficiency and fetal numerical data. No compound-related effects were observed on fertility, implantations, litter size, sex ratio. and pre and post implantation loss.

Table V from the report summarizes resorption data. Treatment had no observed effect on resorptions.

Table VI from the report summarizes external and visceral observations. No treatment-related effects were reported.

Table VII from the report summarizes skeletal variations. No treatment-related effects were reported.

Table VIII from the report is an incidence summary of skeletal variations. No treatment-related effects were reported.

Table IX from the report presents external, visceral and skeletal malformations. No treatment-related effects were reported.

Table I

Mean Body Weights of Pregnant Does[a] During Gestation (kg)

Day	CONTROL Mean	S.E.	N	1.0 mg/kg Mean	S.E.	N	3.0 mg/kg Mean	S.E.	N	9.0 mg/kg Mean	S.E.	N
0	3.01	0.06	16	3.07	0.04	11	3.03	0.04	17	2.99	0.07	13
7	3.10	0.06	16	3.13	0.03	11	3.10	0.04	17	3.09	0.07	13
10	3.13	0.06	16	3.18	0.04	11	3.12	0.04	17	3.09	0.07	13
14	3.19	0.06	16	3.24	0.05	11	3.16	0.04	17	3.09	0.08	13
19	3.24	0.07	16	3.29	0.04	11	3.18	0.05	17	3.10	0.08	13
21	3.25	0.07	16	3.30	0.05	11	3.20	0.05	17	3.09	0.07	13
28	3.33	0.07	16	3.41	0.05	11	3.30	0.05	17	3.22	0.08	13
7-21 GAIN	0.15	0.02	16	0.17	0.02	11	0.10	0.02	17	-0.00**	0.04	13
%GAIN	4.95			5.40			3.15			-0.00		
0-28 GAIN	0.32	0.03	16	0.34	0.04	11	0.27	0.02	17	0.23	0.05	13
%GAIN	10.49			11.24			8.96			7.55		
ACTUAL	2.99	0.08	16	3.04	0.07	11	2.95	0.06	17	2.89	0.09	13
GAIN	-0.03	0.04	16	-0.03	0.05	11	-0.08	0.03	17	-0.11	0.05	13
%GAIN	-0.90			-0.87			-2.62			-3.55		

[a]Includes only does with viable fetuses on Day 28.
**Significantly different from control at the .01 level using Dunnett's test

Table III
Summary of Cholinesterase Data
Mean ± SEM

Gestation Day	Dose mg/kg	N	Plasma IU/l	Plasma % Inhibition	Erythrocyte IU/l	Erythrocyte % Inhibition
20	0	16	400.8 ± 13.4	-	827.2 ± 87.2	-
20	1	13	238.5 ± 12.8*	40.5	249.7 ± 33.8*	69.8
20	3	17	183.1 ± 8.7*	54.3	126.0 ± 29.8*	84.8
20	9	14	131.4 ± 11.0*	67.2	61.5 ± 21.6*	92.6
28	0	16	246.5 ± 12.6	-	910.0 ± 69	-
28	1	12	229.0 ± 26.2	7.5	510.0 ± 83*	44.0
28	3	17	227.4 ± 15.9	7.7	569.0 ± 50*	37.5
28	9	13	213.2 ± 15.7	13.5	544.0 ± 55*	40.2

Gestation Day	Dose mg/kg	N	Brain IU/g	Brain % Inhibition
28	0	16	3224 ± 127	-
28	1	12	3325 ± 160	0
28	3	17	3407 ± 123	0
28	9	13	3080 ± 157	4.5

* Significantly different from control at p less than or equal to 0.05 using Dunnett's test

Table IV

Reproductive Efficiency and Fetal Data

		Control	1.0 mg/kg	3.0 mg/kg	9.0 mg/kg
No. of Pregnant Dams/Total		16/17	15/17	17/17	15/17
Fertility Index[a]		94.1	88.2	100	88.2
Cestation Index[b]		100	93.3	100	100
No. of Litters		16	11	17	13
No. with Resorption Sites Only		0	1	0	0
No. of Deaths Among Dams		0	2	0	1
No. of Dams Which Aborted		0	1	0	1
No. of Corpora Lutea	Median	8.5	8.0	9.0	9.0
	Mean	8.2	7.8	9.1	7.7
	(Range)	(2-12)	(1-11)	(6-13)	(2-13)
Total No. of Implantations		124	91	131	96
	Median	8.0	8.0	9.0	9.0
	Mean	7.8	7.6	7.7	7.4
	(Range)	(2-14)	(1-11)	(3-13)	(1-12)
Total No. of Fetuses		115	85	119	87
Litter Size	Median	7.0	7.5	8.0	8.0
	Mean	7.2	7.1	7.0	6.7
	(Range)	(2-13)	(0-11)	(3-11)	(1-10)
Median Percent Male Fetuses		50.0	40.0	62.5	33.3
Median Wt. Viable Fetuses (gm)	(Male)	34.7	34.3	35.6	36.4
	(Female)	32.6	32.9	34.3	36.0
	(Combined)	34.7	34.1	35.2	35.8
Median Wt. of Placentas		5.1	5.4	5.0	5.6
No. of Resorption Sites	Median	0.0	0.0	0.0	0.0
	Mean	0.6	0.5	0.7	0.7
	(Range)	(0-2)	(0-2)	(0-4)	(0-3)
Total No. of Dead Fetuses		0	1	1	1
	(Range)	(0-0)	(0-1)	(0-1)	(0-1)
% Pre-Implantation Loss	Median	0.0	0.0	0.0	0.0
	Mean	12.2	6.6	20.2	12.4
	(Range)	(0-55.6)	(0-37.5)	(0-76.9)	(0-66.7)
% Post-Implantation Loss	Median	0.0	4.5	0.0	9.1
	Mean	8.3	14.2	8.6	9.6
	(Range)	(0-40.0)	(0-100)	(0-36.4)	(0-28.6)

[a]Fertility Index: ratio of number of pregnant dams/number of dams with successful copulation

[b]Cestation Index: ratio of number of dams with live progeny/number of pregnant dams

Table V

Distribution of Resorptions in Does

No. of Resorptions	Number of Does with Resorptions			
	Control	1.0 mg/kg	3.0 mg/kg	9.0 mg/kg
0	9	7	10	7
1	5	4	4	4
2	2	1	2	1
3	0	0	0	1
4	0	0	1	0
Total No. of Resorptions	9	6	12	9
No. of Does with More than 1 Resorption	2	1	3	2
Percent with Resorptions	43.8	41.7	41.2	46.2

Table VI

External and Visceral Findings on
Fetuses at Termination (Day 28)

Dose[a]	Doe No.	Fetus No.	Observation
Control	RS860	28	Runt (<20.0 g)
	RS885	285	Runt (<20.0 g)
		286	Runt (<20.0 g)
	RS918	353	Cardiovascular anomaly. Aorta ascends straight toward head branching into two carotids; just below where it branches, the aorta arches in an acute fashion dorsally with the right subclavian branching off, aorta then angles acutely to the left beneath the trachea and assumes its normal path
1.0	RS880	119	Non-viable; partially autolyzed but normal for stage of development (10.8 g)
	RS890	309	Left forepaw, downward malrotation
		312	Anterior forepaws, downward malrotation; anophthalmia; nares, abnormal; frontals, pinched together at orbit; testis, small right
		313	Brain, dilated ventricles with fluid
	RS892	315	Runt (<20.0 g)
		317	Runt (<20.0 g)
3.0	RS853	72	Brain, microcephaly with fluid within cranium
	RS898	223	Liver, median lobe, 1 x 1 mm tannish foci, extending into parenchyma
	RS910	365	Adrenal, displaced toward the midline, left
		369	Non-viable; partially autolyzed, normal for stage of development (3.4 g)
9.0	RS863	109	Posterior appendages, inward malrotation, bilateral; tail, kinked; digits, malflexure
	RS872	164	Digit, missing nail 1, left forepaw
		165	Left carotid artery reduced in size, branches off innominate artery opposite right carotid artery
		168	Digit missing nail 1, bilateral forepaw
		169	Digit missing nail 1, bilateral forepaw
	RS877	175	Runt (<20.0 g)
	RS896	265	Ovary, missing left
	RS906	335	Non-viable; partially autolyzed, normal for stage of development (13.2 g)

[a] mg/kg

Table VII

Fetuses with One or More Skeletal Variations

DOSE:	CONTROL		1.0 MG/KG		3.0 MG/KG		9.0 MG/KG	
NO. OF SPECIMENS EXAMINED:	115		84		118		86	
VARIATION AND/OR ABNORMALITY	NO.	%	NO.	%	NO.	%	NO.	%
SKULL: BONES INCOMPLETELY OSSIFIED	17	14.8	13	15.5	12	10.2	13	15.1
SKULL: SUTURES ENLARGED			1	1.2			1	1.2
SKULL: SUTURES FUSED			3	3.6	2	1.7		
SKULL: SUTURES IRREGULARLY SHAPED					1	0.8		
SKULL: FONTANELLE ENLARGED	13	11.3	9	10.7	10	8.5	9	10.5
SKULL: FONTANELLE IRREGULARLY SHAPED					1	0.8		
SKULL: PRESENCE OF CALCIFIED BODY			1	1.2	7*	5.9	2	2.3
SKULL: BONES ABNORMAL			1	1.2	1	0.8		
SKULL: UPPER INCISORS MISSING					1	0.8		
SKULL: IRREGULAR NASAL					1	0.8		
HYOID: VARIATIONS OF HYOID BODY OR ARCH	64	55.7	37	44.0	55	46.6	51	59.3
SKULL: ANGULATED HYOID ARCH			1	1.2	2	1.7	2	2.3
RIBS: EXTRA RIBS	13	11.3	9	10.7	15	12.7	10	11.6
RIBS: INCOMPLETELY OSSIFIED					1	0.8	1	1.2
RIBS: WAVY OR CURVED			1	1.2				
RIBS: BULBOUS OR SPUR			1	1.2				
RIBS: SMALL					1	0.8	1	1.2
RIBS: ABNORMAL POSITION					2	1.7		
RIBS: FLOATING 13TH RIB							2	2.3
VERTEBRAE: CERVICAL-CENTRA INCOMP. OSS.							1	1.2
VERTEBRAE: CERVICAL-CENTRA UNOSSIFIED	1	0.9						
VERTEBRAE: CERVICAL-CENTRA EX OSSIF CNT					1	0.8		
VERTEBRAE: CERVICAL-CENTRA FUSED TO ARCH	1	0.9						
VERTEBRAE: CERVICAL-CENTRA MALPOSITIONED	1	0.9						
VERTEBRAE: CERVICAL-CENTRA IRREGULAR	1	0.9						
VERTEBRAE: THORACIC-CENTRA INCOMP. OSS.					1	0.8		
VERTEBRAE: THORACIC-CENTRA FUSED					1	0.8		
VERTEBRAE: THORACIC-ARCH MALPOSITIONED					1	0.8		
VERTEBRAE: THORACIC-ARCH UNALIGNED					1	0.8		
VERTEBRAE: THORACIC-CENTRA MALPOSITION					1	0.8		
VERTEBRAE: LUMBAR-ARCHES EXTRA	5	4.3	3	3.6	1	0.8	2	2.3
VERTEBRAE: LUMBAR-ARCHES MISSING	1	0.9			1	0.8		
VERTEBRAE: LUMBAR-ARCH UNALIGNED					1	0.8		
VERTEBRAE: SCOLIOSIS					1	0.8		
VERTEBRAE: LUMBAR-CENTRA FUSED					1	0.8		
VERTEBRAE: LUMBAR-CENTRA EXTRA	5	4.3	3	3.6	1	0.8	2	2.3
VERTEBRAE: LUMBAR-CENTRA MISSING	1	0.9						
VERTEBRAE: SACRAL-ARCHES MISSING	5	4.3	3	3.6	1	0.8	2	2.3
VERTEBRAE: SACRAL-ARCHES SHIFT	4	3.5	2	2.4			1	1.2
VERTEBRAE: SACRAL-CENTRA EXTRA	1	0.9						
VERTEBRAE: SACRAL-CENTRA MISSING	4	3.5	3	3.6	1	0.8	2	2.3
VERTEBRAE: CAUDAL-ARCHES FUSED					1	0.8		
VERTEBRAE: CAUDAL-ARCH ABNORMAL			1	1.2	1	0.8		
VERTEBRAE: CAUDAL-ARCH EX OSSIF CNT					1	0.8		
VERTEBRAE: CAUDAL-CENTRA ABNORMAL			1	1.2	1	0.8	1	1.2
VERTEBRAE: CAUDAL-CENTRA INCOMP. OSS.							2	2.3
VERTEBRAE: CAUDAL-CENTRA FUSED					1	0.8		
PELVIS: ILIUM INCOMPLETELY OSSIFIED	1	0.9						
PELVIS: ILIUM UNALIGNED	5	4.3	2	2.4			1	1.2
PELVIS: ISCHIUM-UNALIGNED	1	0.9	2	2.4			1	1.2
PELVIS: PUBIS UNOSSIFIED	1	0.9						
PELVIS: PUBIS INCOMPLETELY OSSIFIED	7	6.1	5	6.0	2	1.7	6	7.0

* Significantly different from control at the 0.05 level

Table VII

Fetuses with One or More Skeletal Variations

DOSE:	CONTROL		1.0 MG/KG		3.0 MG/KG		9.0 MG/KG	
NO. OF SPECIMENS EXAMINED:	115		94		118		86	
VARIATION AND/OR ABNORMALITY	NO.	%	NO.	%	NO.	%	NO.	%
STERNEBRAE: 1ST-INCOMPLETELY OSSIFIED	9	7.8	3	3.6	4	3.4	3	3.5
STERNEBRAE: 1ST-ASYMMETRICAL			1	1.2			1	1.2
STERNEBRAE: 1ST-EXTRA OSSIF CENTER			1	1.2	1	0.8		
STERNEBRAE: 1ST- IRREGULAR	1	0.9	1	1.2	1	0.8	1	1.2
STERNEBRAE: 2ND-INCOMPLETELY OSSIFIED	8	7.0	13	15.5	13	11.0	9	10.5
STERNEBRAE: 2ND-ASYMMETRICAL			1	1.2			1	1.2
STERNEBRAE: 2ND-BIPARTITE			1	1.2				
STERNEBRAE: 3RD-INCOMPLETELY OSSIFIED	1	0.9						
STERNEBRAE: 3RD-ASYMMETRICAL			1	1.2			2	2.3
STERNEBRAE: 3RD- FUSED TO 4TH							1	1.2
STERNEBRAE: 4TH-INCOMPLETELY OSSIFIED	8	7.0	1	1.2	2	1.7	0*	
STERNEBRAE: 4TH-ASYMMETRICAL			1	1.2			2	2.3
STERNEBRAE: 4TH- FUSED TO 5TH	1	0.9	2	2.4			2	2.3
STERNEBRAE: 5TH-UNOSSIFIED	18	15.7	3*	3.6	4**	3.4	2**	2.3
STERNEBRAE: 5TH-INCOMPLETELY OSSIFIED	75	65.2	66	78.6	92	78.0	70*	81.4
STERNEBRAE: 5TH-ASYMMETRICAL			1	1.2			1	1.2
STERNEBRAE: 5TH-BIPARTITE	2	1.7			1	0.8		
STERNEBRAE: 6TH-UNOSSIFIED	3	2.6						
STERNEBRAE: 6TH-INCOMPLETELY OSSIFIED	16	13.9	11	13.3	2**	1.7	14	16.3
STERNEBRAE: 6TH- BIFURCATED PROCESS	2	1.7	1	1.2	1	0.8		
STERNEBRAE: 6TH- IRREGULAR			1	1.2	5	4.2		
SCAPULA: IRREGULAR SPINOUS PROCESS	4	3.5	3	3.6				
CLAVICLE: INCOMPLETELY OSSIFIED	1	0.9						
APPENDAGES: ANTERIOR-UO METACARPALS	9	7.8	10	11.9	3	2.5	9	10.5
APPENDAGES: ANTERIOR-IO METACARPALS	12	10.4	7	8.3	6	5.1	13	15.1
APPENDAGES: ANTERIOR-MISSING METACARP							3	3.5
APPENDAGES: ANTERIOR-UO PHALANGES			4	4.8				
APPENDAGES: ANTERIOR-IO PHALANGES	1	0.9	3	3.6			4	4.7
APPENDAGES: ANTERIOR MISSING PHALANG							3	3.5
APPENDAGES: POSTERIOR-UNOSSIFIED TALUS	4	3.5						
APPENDAGES: POSTERIOR-IO TALUS	7	6.1	2	2.4	4	3.4	4	4.7
APPENDAGES: POSTERIOR-IO METATARSALS	1	0.9						
APPENDAGES: POSTERIOR-UO PHALANGES			2	2.4	1	0.8	2	2.3
APPENDAGES: POSTERIOR-IO PHALANGES	3	2.6	6	7.1			1	1.2

* Significantly different from control at the 0.05 level
** Significantly different from control at the 0.01 level

Table VIII

Incidence Summary of Skeletal
Malformations and Selected Variations

		Litter Incidence (%)	Fetal Incidence (%)
Control	N:	16	115
Malformations		3 (18.8)	3 (2.6)
Variations			
Extra Ribs		6 (37.5)	13 (11.3)
Additional Pre-Sacral Vertebrae		6 (37.5)	9 (7.8)
1.0 mg/kg	N:	11	84
Malformations		2 (18.2)	5 (6.0)
Variations			
Extra Ribs		5 (45.5)	9 (10.7)
Additional Pre-Sacral Vertebrae		3 (27.3)	5 (6.0)
3.0 mg/kg	N:	17	118
Malformations		5 (29.4)	5 (4.2)
Variations			
Extra Ribs		9 (52.9)	15 (12.7)
Additional Pre-Sacral Vertebrae		1 (5.9)	1 (0.8)*
9.0 mg/kg	N:	13	86
Malformations		5 (38.5)	8 (9.3)
Variations			
Extra Ribs		4 (30.8)	10 (11.6)
Additional Pre-Sacral Vertebrae		3 (23.1)	3 (3.5)

* Significantly different from control at the 0.05 level (Fisher's)

Table IX

External, Visceral and Skeletal Malformations

Dose[a]	Doe No.	Fetus No.	Observation
Control	RS857	17	Sternebrae, 4th segment fused to 5th
	RS874	212	Lumbar arch and centra missing
	RS918	353	Cervical centra: fused to arch, malpositioned, irregular; cardiovascular anomaly: aorta ascends straight toward head branching into two carotids, just below where it branches the aorta arches in an acute fashion dorsally with the right subclavian branching off, aorta then angles acutely to the left beneath the trachea and assumes its normal path
1.0	RS890	306	Sternebrae, 4th segment fused to 5th
		312	Skull bones abnormal; sutures fused; caudal arch and centra abnormal; anophthalmia
		313	Sutures fused; brain, dilated ventricles with fluid
	RS897	397	Sutures fused
		398	Sternebrae, 4th segment fused to 5th
3.0	RS853	72	Skull bones abnormal; sutures fused; upper incisors missing; brain, microcephaly with fluid within cranium
	RS879	143	Sutures fused
	RS883	144	Lumbar arch missing; lumbar centra fused; ribs abnormal position; thoracic arch unaligned; lumbar arch unaligned; scoliosis
	RS901	233	Caudal arch and centra fused, abnormal
	RS908	363	Thoracic centra: fused, malpositioned; thoracic arch malpositioned; ribs abnormal position
9.0	RS863	159	Caudal centra abnormal; tail kinked
	RS872	164	Digit: missing nail 1; metacarpal and phalange missing
		165	Cardiovascular anomaly: left carotid reduced in size, branches off innominate artery opposite right carotid artery
		168	Digit, missing nail 1; metacarpal and phalange missing
		169	Digit, missing nail 1; metacarpal and phalange missing
	RS877	171	Sternebrae, 4th segment fused to 5th
	RS896	265	Ovary, missing left
	RS899	268	Sternebrae, 3rd segment fused to 4th, 4th segment fused to 5th

[a] mg/kg

Tox Chem No. Tribufos (DEF) File Last Updated _____ Current Date _____

Study/Lab/Study #/Date	Material	EPA MRID No.	Results: LD_{50}, LC_{50}, PIS, NOEL, LEL	TOX Category	CORE Grade/ Doc. No.
Onco-mouse; Mobay; 86-271-01	Tech 98.9%	411710-01	Doses tested 0, 10, 50 or 250 ppm for 90 weeks. At 10 ppm, decreased plasma and RBC cholinesterase both sexes, decreased brain cholinesterase males, at 78 weeks males decreased MCV and MCH, at week 90 females decreased hematocrite. At 50 ppm, males increased number showing paleness and hunched backs, at 78 weeks males decreased MCV and MCH, at week 90 decreased MCH, at week 90 females decreased RBC count, hemoglobin and hematocrite. Histopathology males; adrenals amyloid, epididymis hyperspermatogenensis, small intestine amyloid and vacuolar degeneration epithelium, spleen hematopoiesis. At 250 ppm loose stools females, enlarged abdomen both sexes, increased mortality/decreased life span both sexes, increased food consumption and body weight both sexes, decreased RCB count, hemoglobin, hematocrite, MCV and MCH in males, decreased RCB count, hemoglobin and hematocrite in females. Histopathology males, adrenals degeneration, liver hemangiosarcoma*, rectum acute inflammation, necrosis and ulcer, small intestine adenocarcinoma*, dilated/ distended and mucosal hyperplasis. In females, adrenals calcification and degeneration/ pigmentation, caecum edema, liver hypertropy, lung alveolar/bronchiolar adenoma*, mesenteric lymph node congestion, rectum acute inflamation, necrosis and ulcer, small intestine adenocarcinoma*, dilated/distended, mucosal hyperplasia.	N/A	Guideline

Tox Chem No. Tribufos (DEF) File Last Updated _____ Current Date _____

Study/Lab/Study #/Date	Material	EPA MRID No.	Results: LD_{50}, LC_{50}, PIS, NOEL, LEL	TOX Category	CORE Grade/ Doc. No.
Teratology-rat; Miles Laboratories: 87320: 8/8/86	Tech 98%	401906-01	Pregnant rats were dose orally at 0, 1, 7 and 28 mg/kg/day (days 6-16). Maternal RBC and plasma cholinesterase activity was depressed at 7 and 28 mg/kg/day and brain activity at 28 mg/kg/day. Maternal weight gain was decreased at 28 mg/kg/day. Maternal toxicity LEL 7 mg/kg/day, NOEL 1 mg/kg/day. Fetotoxic NOEL 28 mg/kg/day (HDT).	N/A	Guideline
Teratology-Rabbit; Miles Laboratories: MTD0003; 1/22/87	Tech 98%	401906-02	Pregnant rabbits were dosed at 0, 1, 3 or 9 mg/kg/day, days 7-19. Plasma and RBC cholinesterase activity was significantly reduced at all doses on day 20 and RBC at all doses on day 28. Does failed to gain weight at 9 mg/kg/day during dosing. Maternal toxicity LEL 1 mg/kg/day (LDT). Fetal toxicity NOEL 9 mg/kg/day (HDT).	N/A	Guideline