Microfiche

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

009870

DEC 0 1 1992

OFFICE OF
PREVENTION, PESTICIDES
AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: EPA Id. No.: 053301. Fenthion: Review of the rat carcinogenicity/chronic feeding study submitted in 1991.

TOX CHEM No.: 456F
PC No.: 053301
TOX PROJECT Nos.: 1-1837 / D166778
Submission Nos.: S399913

FROM: John Doherty, PhD. 11/13/92
Section IV, Toxicology Branch I
Health Effects Division (H7509C)

TO: Larry Schnaubelt/Richard King
Product Manager #72
Special Review and Reregistration Division
(H7505C)

THROUGH: Marion Copley, DVM, Section Head 11/16/92
Section IV, Toxicology Branch I
Health Effects Division (H7509C)

KB 11/19/92

I. CONCLUSION

The rat chronic feeding/oncogenicity study was reviewed and the chronic feeding aspect of the study determined to be CORE MINIMUM. Since there was minimum to moderate inhibition of plasma ChE and RBC and brain AChE, no NOEL was established for these parameters. Since TB-I considers that the resolving power of the enzyme assay would not distinguish the potential inhibition at dose levels only a few ppm lower than the 5 ppm already tested, classification of this study as MINIMUM is considered to be appropriate. NOEL/LELs were established for ophthalmological and other systemic lesions. The carcinogenicity aspect of the study was determined to be CORE GUIDELINE. No evidence of carcinogenicity was apparent.

No additional series 83-1, 83-3 or 83-5 studies with rats are required at this time. Additional data, however, may be required to further understand the onset and nature of the optic pathology of fenthion in rats. This requirement (if any) will be determined at a later time under a separate cover.

II. ACTION REQUESTED

The Mobay Chemical Company has submitted a rat carcinogenicity/chronic feeding study (series 83-5) in response to reregistration requirements. This study has been reviewed and the following comments apply.

III. Toxicology Branch Comments. Refer identification of the study and results summary under item 7 below and to the DER attached.

1. Carcinogenicity. There was no evidence to indicate a concern for carcinogenicity at dose levels up to and including 100 ppm.

2. ChE and AChE inhibition. No NOEL was established for inhibition of plasma ChE, RBC and brain AChE. At the lowest dose level tested there was 31-41% in females and 7-27% in males inhibition of plasma ChE throughout the study; 7-18% in females and 1-16% inhibition of RBC AChE throughout the study and 13-14% inhibition of brain AChE in males at termination.

TB-I does not at this time recommend that additional testing be required to establish the NOEL/LEL for fenthion on ChE and AChE in the rat. The low level of (but consistent) inhibition noted at the lowest test dose level in this study would not justify the expense of requiring additional chronic feeding data. TB-I considers that the resolving power of the enzyme assay would not distinguish the potential inhibition at dose levels only a few ppm lower than the 5 ppm already tested.

The RfD committee may decide the rat species is critical in setting the NOEL for the RfD and that additional data are necessary for this purpose. Alternatively, the RfD committee may decide to use an additional modification factor to account for the lack of a NOEL. The decision not to require additional data for ChE/AChE may be reevaluated at that time.

3. Effects on the rat eye and optic nerve. The eye and/or optic nerve were assessed by ophthalmoscopy, electroretinogram (ERG) and pathology. Each of these investigational endpoints indicated

that fenthion affected the visual system. In general, females were more susceptible than males. The NOEL/LEL was established at 5/20 ppm based primarily on the ERG data. Refer to the DER for details of the lesions/effects produced by fenthion.

Lesions in the eye and optic nerve were most evident at terminal sacrifice. Investigations before that time did not yield strong evidence for effects.

The effects on the optic nerve (atrophy) were considered to be in females only in the high dose group and only 5 were affected. Only those animals with bilateral atrophy were considered affected by the test material.

In general, a decision on the potential effects of fenthion on the eye and optic nerve will not be based on this study alone. Additional data may be requested pending resolution within the Agency of policy regarding this special type of toxicity problem for organophosphates.

4. Epididymal pathology. Vacuolation of the epididymides was demonstrated to have a NOEL/LEL of 5/20 ppm in the rat chronic feeding study. This finding is consistent with the similar observation noted in the rat multi generation reproduction study (MRID No.: 413486-01, HED Document No.: 8545) which demonstrated a NOEL/LEL of 2/14 ppm. It should be noted that each study utilized a different strain of rat. The rat multi generation reproduction study indicated a NOEL/LEL of 14/100 ppm for decreased fertility. This may have been related to the epididymal vacuolation.

5. Nasolacrimal duct pathology and lung weight and "inhalation pneumonia".

Pathological findings in the nasolacrimal duct and "inhalation" pneumonia were noted in both males and females NOEL/LEL = 5/20 ppm). The study report implied that these conditions may be secondary effects of ChE/AChE inhibition. In particular, the excess secretions due to the inhibition causes the accumulation and subsequent pneumonia. TB-I considers this a novel explanation but within the experiences of TB reviewers, similar findings have not been a consistent finding for other organophosphates and carbamates. Thus, until further notice these conditions are considered to be effects of fenthion.

6. Mineralization.

Mineralization of the stomach was noted in the high

dose (100 ppm) group males and females. This condition was also said by the study author to be related to ChE/AChE inhibition but it has not been noted with other organophosphates and carbamates.

7. Study reviewed.

Study Identification/ Classification	TB Comments and NOEL/LELs
83-5. Chronic Feeding/Carcinogenicity - rats. Mobay Corporation Study No.: 87-271-01. December 17, 1990. MRID No.: 417431-01 (5 volumes). Classification Chronic Feeding Aspect: MINIMUM Carcinogenicity Aspect: GUIDELINE	**LEL (ChE and AChE):** 5 ppm. NOEL not established. Moderate inhibition of plasma ChE (40%) and minimal inhibition of RBC (up to 19% and brain (14%) AChE at this level. (Testing at lower levels not required at This Time) **NOEL/LEL (Systemic effects):** 5/20 ppm. At 20 ppm: epididymal pathology; vacuolation of nasolacrimal duct (F); pneumonia (M); lung weight (M); skin lesions and clinical signs (threshold, M/F). At 100 ppm: Body weight decreases in adults (M/F), mineralization (stomach and other structures); vacuolation of nasolacrimal duct (M). **NOEL/LEL (Eye and optic nerve effects):** 5/20 ppm. At 20 ppm. Eye function and/or pathology (F): At 100 ppm: Additional eye and optic nerve pathology (atrophy and/or neovascularization, M/F). Refer to DER for details. **Dose levels tested:** 0, 5, 20 and 100 ppm corresponding to 0, 0.09, 0.3 and 1.8 mg/kg/day for males and 0.07, 0.3 and 1.5 mg/kg/day for females. Fischer 344 strain rat.

[83-5. Fenthion/1992]

Reviewed by: John Doherty, Ph.D. 11/13/92 009870
Section IV, Tox. Branch (H7509C)
Secondary reviewer: Marion Copley, DVM
Section IV, Tox. Branch (H7509C) 11/14/92

DATA EVALUATION REPORT

STUDY TYPE: 83-5. Chronic feeding/oncogenicity - Rats

Tox Chem No.: 456F PC. No.: 053301

MRID NUMBER: 417431-01 (five volumes) and 424572-01 (Supplementary report)

TEST MATERIAL: Fenthion (see description below).

STUDY NUMBER(S): 87-271-01

SPONSOR: Mobay Corporation

TESTING FACILITY: Mobay Corporation, Stilwell, Kansas

TITLE OF REPORT: "Combined Chronic Toxicity/Oncogenicity Study of Technical Grade Fenthion (Baytex) with Rats".

AUTHOR(S): W.R. Christenson

REPORT ISSUED: December 17, 1990
Study Dates: January 19, 1987 to January 31, 1989.

CONCLUSIONS:

LEL (plasma ChE and RBC and brain AChE): 5 ppm. NOEL not established. Moderate inhibition of plasma ChE (40%) and minimal inhibition of RBC (up to 19% and brain (14%) AChE at this level (testing at lower levels not required at this time - see result: Clin. Chem.)

NOEL/LEL (Systemic effects): 5/20 ppm. At 20 ppm: epididymal pathology; vacuolation of nasolacrimal duct (F); pneumonia (M); lung weight (M); skin lesions and clinical signs (threshold, M/F). At 100 ppm: Body weight decreases in adults (M/F), mineralization (stomach and other structures); vacuolation of nasolacrimal duct (M).

NOEL/LEL (Eye and optic nerve effects): 5/20 ppm. At 20 ppm. Eye function and/or pathology (F): At 100 ppm: Additional eye and pathology (atrophy and/or neovascularization, M/F), optic nerve degeneration (F). Refer to DER for details.

Carcinogenicity: No evidence of carcinogenicity potential.

Dose levels tested: 0, 5, 20 and 100 ppm corresponding to 0, 0.09, 0.3 and 1.8 mg/kg/day for males and 0.07, 0.3 and 1.5 mg/kg/day for females. Fischer 344 strain rat.

Classification: Chronic feeding study: Core-MINIMUM. Carcinogenicity study: GUIDELINE. The study satisfies the

requirement for a 83-5 combined chronic feeding/carcinogenicity study or 83-1 chronic feeding study in rodents and an 83-2 oncogenicity study in rats.

Quality assurance statement was provided and signed by C.A. Halder on Dec. 14, 1990 (some 86 reports to the study director or management were filed).

Good Laboratory Practice Compliance Statement was provided and signed by the submitter, sponsor and study director.

REVIEW

A. MATERIALS:

1. Test compound: Fenthion (O,O-dimethyl O-[3-methyl-4-(methylthio)phenyl]phosphorothioate), an organophosphate insecticide also know as Baytex.

From Batch #85R-01-461. Analytical results indicated test material was 94.8% pure and not 97.0% as indicated in the test substance identification sheet. The impurities consisted of [redacted] components were present at 0.01 (?, probably a misprint mean to be 0.1) to 1%, [redacted] were present at 0.01 to 0.1% and the remainder were present at < 0.01%.

2. Test animals: Species: rat, Strain: Fischer 344 (CDF(F-344)/Crl/Br). Source: Charles River Laboratories, Inc. Kingston, N.Y. The report asserts that the rats were not greater than six weeks of age when placed on the study.

B. STUDY DESIGN:

1. Animals were assigned by computer assisted randomization to the following test groups:

Table 1. Experimental Design.

Dose Group	Compound Level (ppm)	Main Study (24 months) Number of rats Males	Females	Interim Sacrifice (12 months) Number of rats Males	Females	Compound Intake (mg/kg/day) Males	Females
1 Control	0	50	50	20	20	0.0	0.0
2 Low (LDT)	5	50	50	-	-	0.09	0.07
3 Mid (MDT)	20	50	50	-	-	0.3	0.3
4 High(HDT)	100	50	50	20	20	1.8	1.5

2. Diet preparation

Fenthion was prepared in corn oil and admixed into the feed (Purina Mills Rodent Chow 5001-4) weekly and stored at -23°C. A given batch of feed was reportedly available for 1 week.

Samples of treated food were analyzed for homogeneity, stability and concentration.

A. Homogeneity. Three samples for each of the 5 and 100 ppm test diets were reportedly taken from the top middle and bottom of the mixing bowl and analyzed. The 5 ppm diet resulted in a mean value of 4.5 ± 0.19 (coefficient of variation or CV 4%) and it was apparent that each layer had the same level of fenthion.

The 100 ppm diet had only 82-87 ppm in the top layer while there was 92 to 97 ppm in the middle layer and 91 to 106 ppm of fenthion in the bottom layer. These data indicated a mean of 92.6 ± 7.5 ppm (CV 8% and 93% of the target). Since other mixing samples were not assessed for homogeneity it is not possible to tell if all preparations had this poor uniformity of mixing. Some of the samples may have been off by 20%. This, however, would still give a dietary level substantially higher than the next lower dose level of 20 ppm.

B. Stability. Samples of the 5 and 100 ppm test diets were stored at room temperature for 14 days and assessed for fenthion content on days 0, 1, 3, 7, 9 and 14. The 5 ppm test diet dropped from 100% to 82% by day 7 and was still 82% at day 14. The 100 ppm test diet dropped from 100% to 95% by day 7 and to 87% by day 14. The readings at day 9 for both test diets were lower than for day 14. Since the test diets were changed weekly, TB-I is not critically concerned with the stability data after day 7. Fenthion is considered to be sufficiently stable within 7 days so as to not compromise the study.

C. Concentration. The results of eleven preparations of fenthion diets gave means of 4.8 ± 0.55 (CV 12%), 17.9 ± 1.5 (CV 8%) and 92.5 ± 6.7 (CV 7%) to indicate that the 5, 20 and 100 ppm diets were 96, 90 and 92% of their nominal concentrations. This is considered within acceptable limits.

4. Statistics - The following procedures were utilized in analyzing the numerical data:

Table 2. Statistical methods used.

Data	Statistical test(s)
Continuous (except organ weight)	ANOVA followed by Duncan's Multiple Range Test for parameters which showed a significant F value in ANOVA.
Organ weights	Bartlett's test for equality of variance. Group means with equal variance were further analyzed by ANOVA followed by either Dunnett's or Mann Whitney U-Test for between group comparisons if a significant F value was obtained by ANOVA.

Frequency data	Chi-square followed by Fisher's Exact Test (one-tailed in cases of significant variation by the Chi-square analysis,

The report states that "when deemed appropriate, additional statistical tests to analyze continuous and frequency data were used".

The statistical evaluations were reportedly performed using software from either INSTEM Computer Systems, Stone, Staffordshire, England of SAS Institute inc., Cary, North Carolina. The level of significance was set at $p \leq 0.05$.

C. **METHODS AND RESULTS:**

1. Observations

Animals were reportedly inspected twice daily (but once on weekends and holidays) for signs of toxicity and mortality. More detailed physical examinations for clinical signs of toxicity and palpation for masses were made weekly.

Toxicity (clinical signs): The report asserts that clinical signs were increased in the male and female high dose groups. These signs included eye opacity zones (M), urine stains(M), alopecia(M), hunched back, loose stool(M), rough coat, enlarged preputial gland (M) and "irritation of the penis", lesions of the tail and posterior paws. [M = The mid dose group may also have been affected based on the number of animals affected.] In general many of these conditions were more frequent in females.

CONCLUSION (Clinical signs): NOEL = 5/20 ppm. TB-I acknowledges that 20 ppm is likely the threshold dose level

Mortality (survival). There was greater than 50% survival for both sexes at study termination. Survival among the male groups was not affected. There were 29, 29, 25 and 31 (of 50) males surviving to termination for the control to high dose groups respectively.

The study report maintained that survival among the females was not affected although there were 35, 36, 33 and 29 females surviving to termination. These data indicate a possible effect on survival in the high dose group. This possibility is again reflected when the mean time to death is considered which was 697, 704, 701 and 644 days for the control, low, mid and high dose groups respectively. There was, however, no common cause of death listed and the clinical symptoms in females do not suggest

severe toxicity.

009870

CONCLUSION (Survival): NOEL > 100 ppm.

2. Body weight

Animals were reportedly weighed weekly for the entire study. The study report maintains that there were no effects on the weight or weight gain of either males or females except at the high (100 ppm) dose level. Table 2 illustrates body weight data at selected intervals for both males and females.

Table 3. Body weight and/or gain at selected intervals.

Week Interval		Dose Level[1,3] Control	5 ppm	20 ppm	100 ppm
1	M	107.5	110.5	114.9*	122.3*
	F	91.8	95.3*	98.2*	99.7*
13	M	303.4	308.4	305.3	303.0
G[2]		195.9	197.9	190.4 (-3%)	180.7 (-8%)
	F	177.3	177.6	176.2	174.2
G[2]		85.5	82.3	78.0 (-9%)	74.5 (-13%)
26	M	350.1	352.4	352.6	340.9*(-3%)
	F	193.0	195.0	197.7*	186.0*(-4%)
52	M	401.0	402.0	385.4	380.3*(-5%)
	F	223.4	229.2	225.9	210.6*(-6%)
T	M	368.0	366.1	366.3	338.7*(-8%)
	F	264.7	270.3	269.7	238.3*(-10%)

* Statistically significantly different from control $p < 0.01$.
1. Data are the mean weight based on number of animals surviving at the interval.
2. For week 13, the main growth period the weight gain (g) is given. and the % gain relative to the controls is in ().
3. The start of the study was staggered resulting in the apparent increase in body weight as shown (see conclusions for discussion).

Initial response. The weights of the rats were reportedly equal for all groups at initiation but at week one there was an apparent increase and dose response for all groups for females but only for the mid and high dose groups for males. The statistical increase disappeared after week four or five. This apparent increase is not considered to be compound related but to relate to the staggering of the starting times of dosing.

Growth period (weeks 0-13). The initial increase in weight obscures the weight gain pattern for this period. There was a slight decrease noted for both the male and female high

dose group but statistical significance was not demonstrated. The body weights for males and females were essentially equal at week 13.

Mature animals (week 13 to termination). The high dose group males were statistically significantly lower (most often <10%) from week 19 to week 104. The high dose females were lower from weeks 16 to termination. The middle dose was occasionally statistically significantly higher.

CONCLUSION (Body weight): NOEL/LEL = 20/100 ppm. At 100 ppm: Male and female body weight decreases in adults.

[NOTE: TB-I recognizes an apparent initial weight gain noted at all dose levels in females and in the mid and high dose level in males but accepts the registrant's explanation this apparent effect is related to the staggering of the start of dosing (refer to Supplemental Submission under MRID No.: 424572-01 dated August 13, 1992). In addition, the apparent weight effect is not supported by accompanying clinical signs and is reversible.

3. Food consumption and compound intake. Food consumption was measured weekly.

There were no compound related effects on food consumption reported. Refer to Table 1 for calculated compound intake.

4. Ophthalmological examinations. All ophthalmological and other data related to the eye and optic nerve are discussed in the pathology Discussion (Section 9A) below.

Note: The summary table and the methods section do not indicate the units of measure for the hematology, clinical chemistry and urinalysis data. As per supplementary response provided by the company, this information is on pages 2355 and 2691. A copy of the listing of the parameters investigated and the units of measure is attached to this DER.

5. Hematology and Clinical Chemistry. Blood was collected at 3, 6, 12 and 18 and at 24 months for hematology and clinical analysis from the orbital sinus after the rats were fasted overnight from 20 rats/sex/dose group.

The CHECKED (X) parameters were examined.

5a. Hematology

X		X	
x	Hematocrit (HCT)*	x	Leukocyte differential count*
x	Hemoglobin (HGB)*	x	Mean corpuscular HGB (MCH)
x	Leukocyte count (WBC)*	x	Mean corpuscular HGB conc.(MCHC)

x	Erythrocyte count (RBC)*	x	Mean corpuscular volume (MCV)
x	Platelet count*	x	Reticulocyte count
	Blood Clotting Measurements		
	(Thromboplastin time)		
	(Clotting time)		
	(Prothrombin time)		

009870

* Required for subchronic and chronic studies

There were no consistent dose related effects on any of the hematological parameters investigated.

5b. Clinical Chemistry

Electrolytes:		Other:	
x	Calcium*	x	Albumin*
x	Chloride*	x	Blood creatinine*
	Magnesium*	x	Blood urea nitrogen*
x	Phosphorous*	x	Cholesterol*
x	Potassium*	x	Globulins
x	Sodium*	x	Glucose*
		x	Total Bilirubin*
		x	Total Serum Protein*
x	Triglycerides		
			Serum protein electrophoresis

Enzymes

x	Alkaline phosphatase
x	Creatinine phosphokinase*
x	Lactic acid dehydrogenase
x	Serum alanine aminotransferase (also SGPT)*
x	Serum aspartate aminotransferase (also SGCT)*
x	gamma glutamyl transferase
	glutamate dehydrogenase
x	plasma ChE, RBC AChE and brain (collected at necropsy) AChE.

* Required for subchronic and chronic studies

5b1. Ca++. The pathology report identified several organs with an apparent compound related increase in mineralization. This condition might be related to an effect (indirect or direct) on Ca++ metabolism that would in turn be reflected in the blood Ca++ levels.

Ca++ levels (in mg/dl) for males were 11.2 for the control and 10.9 (2.7%), 10.8 (3.6%) and 10.7 (4.5%) decreased at week 27 for the low, mid and high dose group respectively. Ca++ levels remained lower at week 53 being 11.9, 11.5 (3.4%), 11.3 (5.0%), and 11.4 (4.2%). They were not statistically decreased at weeks 78 and 105. Female Ca++ levels were also slightly lower for the high dose group at week 27 (4.5% lower), for the mid (3.4% lower) and high (2.5% lower for the main group and 5.2% for the satellite group) at week 53. The high dose group was slightly elevated at week 105 (3.5% higher). TB-I does not consider that these fluctuations in Ca++ level are of a toxicological concern. They are considered too minor to account for the mineralization noted in the pathology of certain organs. It would also be expected that if changes in Ca++ levels were

associated with the mineralization, these changes would more likely occur in the later weeks of the study rather than at weeks 27 and 53 as noted.

5b2. Other parameters. The study report notes that serum glucose was reduced for females during weeks 14 to 79 for the mid (about 8%) and high (about 14%) dose groups. Male glucose levels were also occasionally reduced (and in some instances elevated). BUN, Cl-, triglycerides, cholesterol, creatinine phosphokinase, protein, albumin, globulin, phosphate, K+ and uric acid all showed occasional significant changes from the control. Because of the low magnitude of the difference, a lack of consistency in the direction and/or persistence with time, TB-I does not consider these differences toxicologically relevant.

NOEL (Clinical chemistry) > 100 ppm (HDT).

5b3. Plasma ChE, RBC AChE and Brain AChE.

Blood was obtained from the orbital sinus at 3, 6, 12, 18 and 24 months. The plasma ChE and RBC AChE were assessed by the method of Hackathorn (American Industrial Hygiene Association Journal 44:547 (1983), paper entitled "Validation of a Whcle Blood Method for Cholinesterase Monitoring"). The method to assess for brain AChE was not specifically stated.

Table 4A. Plasma ChE (in U/ml) activity and inhibition data.

Month		Control	5 ppm	20 ppm	100 ppm
3	M	0.53	0.46*(13.2%)	0.38*(28.3%)	0.31*(41.5%)
	F	2.42	1.42*(41.3%)	0.78*(67.8%)	0.52*(78.5%)
6	M	0.61	0.51*(16.4%)	0.40*(34.4%)	0.31*(49.2%)
	F	2.53	1.56*(38.3%)	0.80*(68.4%)	0.53*(79.1%)
12	M	0.68	0.63*(7.4%)	0.47*(30.9%)	0.37*(45.6%)
	F	2.80	1.81*(35.4%)	0.95*(66.1%)	0.59*(78.9%)
18	M	0.98	0.79*(19.4%)	0.51*(48%)	0.35*(64.3%)
	F	2.61	1.64*(37.2%)	0.88*(66.3%)	0.55*(78.9%)
24	M	1.27	0.92*(27.6%)	0.53*(58.3%)	0.39*(69.3%)
	F	2.32	1.60*(31.0%)	0.76*(69.8%)	0.52*(77.6%)

* $p < 0.05$, study report statistics. Note: No other levels of statistical significance were reported.

The above table shows that male plasma ChE was inhibited 7.4 to 27.6% at the 5 ppm dose group throughout the study. The mid (28.3 to 58.3%) and high (41.5 to 69.3%) levels were inhibited to a progressively greater extent.

Female plasma CHE was inhibited at the 5 ppm dose level 31.0 to 41.3% throughout the study. The mid (66.1 to 69.8%) and high (77.6 to 79.1%) dose levels were inhibited to a progressively greater extent.

Table 4B. RBC (in U/ml) and brain AChE (in U/gm) data and inhibition.

Month		Control	5 ppm	20 ppm	100 ppm
3	M	2.80	2.51*(10.4%)	1.93*(31.1%)	1.41*(49.6%)
	F	2.69	2.38*(11.5%)	1.75*(34.9%)	1.49*(44.6%)
6	M	2.94	2.72*(7.5%)	1.98*(32.7%)	1.47*(50%)
	F	2.75	2.52*(8.4%)	1.82*(33.8%)	1.43*(48%)
12	M	2.67	2.64(1.1%)	1.92*(28.1%)	1.25*(53%)
	F	2.53	2.30*(5.9%)	1.65*(34.8%)	1.19*(53%)
18	M	2.87	2.40*(16.4%)	1.85*(35.5%)	1.21*(57.8%)
	F	2.70	2.20*(18.5%)	1.61*(40.4%)	1.21*(55%)
24	M	3.13	3.05(2.6%)	2.63*(16%)	1.94*(38%)
	F	3.16	2.94*(6.9%)	2.26*(28.5%)	1.86*(41.1)
Brain AChE	M	13.6	11.8*(13%)	8,2*(39.2%)	3.3*(76%)
	F	13.9	11.9*(14.1)	7.9*(43.4%)	3.0*(78.3%)

* $p < 0.05$. Study report statistics. Note: No other levels of statistical significance were reported.

The above table shows that male RBC AChE was significantly inhibited (7.5 to 16.4%) in three of the five assay intervals. The mid (16 to 35.3%) and high (38 to 57.8%) dose levels were inhibited to a progressively greater extent.

Female RBC AChE was inhibited 5.9 to 18.5% in the low dose (5 ppm) group. The mid (28.5 to 40.4%) and high (41.1 to 55%) dose levels were inhibited to a progressively greater extent.

Both male (13%) and female (14%) brain AChE was inhibited in the low (5 ppm) dose group. The mid (39.2% for males and 43.4% for females) and high (76% for males and 78.3% for females) dose levels were inhibited to a progressively greater extent.

CONCLUSION (ChE and AChE): LEL = 5 ppm for all three enzyme sources (plasma, RBC and brain). NOEL not established.

Note: TB-considers that the consistency of the inhibitory effect is more important than the fact that inhibition did not exceed 20%. There is also good dose response at the higher dose levels to support the conclusicn that 5

ppm is a significant dose level for inhibition of these enzymes. Thus, the LEL is set at lower dose levels than which the study authors set. TB-I considers that the type of testing at lower dose levels to establish more definite NOEL/LELs is limited by the sensitivity of the assay methods and their power of resolution. It is unlikely that the resolving power of the enzyme assay would distinguish potential inhibition at dose levels much lower than the low dose already tested (5 ppm).

6. Urinalysis

Urine was collected from fasted animals at months. The CHECKED (X) parameters were examined.

[x]	Appearance*	[x]	Glucose*
[ ]	Volume*	[x]	Ketones*
[x]	Specific gravity*	[x]	Bilirubin*
[x]	pH	[x]	Blood*
[x]	Sediment (microscopic)*	[ ]	Nitrate
[x]	Protein*	[x]	Urobilinogen

There were no consistent dose related effects on any of the urinary parameters investigated.

CONCLUSION (hematology, clinical chemistry (except CHE/AChE), urinalysis except ChE and AChE): NOEL > 100 ppm.

7. Organ weight. The following organs were weighed and absolute and relative to body weighs were reported: adrenals, brain, heart, kidneys, liver, lungs, ovaries, spleen and testicles. Data were presented for the terminal sacrifice group only[1]. There was no evidence presented that the interim sacrifice group had organ weight determinations. The following were noted:

-Lungs: absolute weight at termination was decreased ($p < 0.05$) 15% (mid dose) and 18% (high dose) for males, no decrease in females.

relative weight at termination was not statistically decreased but was 12 to 14 % decreased for the high and mid dose groups for males respectively.

-Brain: elevated 2% ($p < 0.05$) for both the mid and high dose males but no dose response, no increase in females.

---

[1]The Table for both relative and absolute organ weight lists "day 97" for the interval. The numbers of organs weighed correspond to the survivors after 2 years, thus TB-I concludes that the entries are for the groups surviving for two years. The term "97" is a computer error. Refer to Supplementary report provided by the registrant (MRID No.: 414572-01).

-Liver: decreased ($p < 0.05$) 11% for male high dose group and 8% for the female high dose group.

-Kidney: decreased ($p < 0.05$) 5% for the female high dose group, no decrease in males.

-Testis: high dose group decreased 15.2% but not significant. There was a very large (40%) standard deviation for the testis data.

[Note: The epididymides was not weighed.]

CONCLUSION (Organ Weights): NOEL/LEL = 5/20 ppm. At 20 and 100 ppm lung weight decreased. This weight effect may be associated with "pneumonia". See pathology section.

Note: The other organ weight changes are considered to be correlated with the overall body weight change. These organs will be commented on further in the pathology section.

8. Sacrifice and Pathology -

All animals that died and that were sacrificed (by CO_2) on schedule were subject to gross pathological examination and the CHECKED (X) tissues were collected for histological examination. Standard preparation and staining procedures were employed. The rats that were sacrificed in extremis and at scheduled termination had their eyes, Harderian gland and optic nerve placed in universal fixative. The rats which were found dead had these structures placed in 10% formalin.

No evidence that any special stains were utilized was presented. The pathology report (Appendix VIII of the study was authored by S.G. Lake). The pathology data were peer reviewed by an independent pathologist. The following organs/tissues were reportedly assessed.

	Digestive system
	Tongue
x	Salivary glands
x	Esophagus
x	Stomach
	Duodenum*
	Jejunum*
	Ileum*
x	Cecum
x	Colon
x	Rectum
x	Liver
	Gall bladder
x	Pancreas

	Respiratory
x	Trachea
x	Lung
	Nose

	Cardiovasc./Hemat.
x	Aorta
x	Heart
x	Bone marrow
x	Lymph nodes
x	Spleen
x	Thymus

	Urogenital
x	Kidneys
x	Urinary bladder
x	Testes
x	Epididymides
x	Prostate
x	Seminal vesicle
x	Ovaries
x	Uterus

	Neurologic
x	Brain
x	Periph. nerve
x	Spinal cord
x	Pituitary
x	Eyes (optic n.)

x	Adrenals
	Lacrimal gland
x	Mammary gland
x	Parathyroids
x	Thyroids

	Other
x	Bone (femur, rib, sternum)
x	Skeletal muscle
x	Skin (hindlimb, tail, ear)
	All gross lesions and masses

| | Pharynx
|x| Larynx

*small intestine is listed but not the individual subsections.

Others (most very limited number of animals examined): adipose tissue, blood vessel, cervix, clitoral gland, ear, Harderian gland, femorotibial joint, lymph nodes (other, cervical, mesenteric), mesentery, muscle (other), oviduct, penis, prepuce, preputial gland, scrotum, skin (other), skull, and vagina.

Individual organ/tissue discussions. [Note The pathology summary tables (MF1-SUM) did not indicate which lesions were statistically significantly increased.]

9A. **Assessment of the visual system and associated structures.** Fenthion has been implicated as having an adverse effect on the eye and optic nerve. This study included both conventional and special assessments for determining if fenthion affected the visual system. The following is an outline of the methods and results used to assess for effects on the eye, optic nerve and vision.

1. Ophthalmology. Assessments were reportedly made prior to initiation of dosing and prior to sacrifice on all animals. Animals showing eye lesions in the pre-study assessment were removed from the study. Three different investigators made the assessments: Dr. R.H. Hayes did the pre-study exam, Dr. H.D. Hoang did the interim sacrifice group and Dr. L.H. Rubin (professor of Veterinary Ophthalmology at U. Pennsylvania) did the terminal ophthalmology exam. The assessment included:

Part I: Initially examined prior to dilation with (Mydriacyl 1%):

Pupil reflex - using light reflected from the viewing mirrors of an ophthalmoscope.

Conjunctiva, cornea and iris - examined grossly and by use of a slit lamp microscope or ophthalmoscope.

Part II. Examined following mydriasis:

Lens, vitreous humor and retina were examined with an ophthalmoscope.

An indirect method was used to evaluate the eyes by ophthalmoscopy. This method used a condensing lens (+20D or +28D) placed between the eye and an ophthalmoscope.

Results:

There were no significant ophthalmoscopic findings

reported in the rats examined during study week 75 on the rats that were to be used for the electroretinogram (ERG) assessment (see below). This was a limited assessment and only the left eye was examined.

At termination there was evidence of effects of fenthion on the eye. Table 5 illustrates these findings.

Table 5. Adverse effects noted at ophthalmoscopy.

Lesion	Sex	Dose Level: Control	5 ppm	20 ppm	100 ppm
Diffuse posterior subcapsular cataract	M	2/32	1/29	2/29	1/34
	F	1/37	1/36	1/38	14/32 (i)
Possible[1] retinal degeneration	M	0/32	0/29	0/29	0/34
	F	0/37	1/36	0/38	26/32 (i)
Phthisis bulbi	M	0/32	2/29	0/29	2/34
	F	1/37	1/36	1/38	4/32 (i)
Synechia anterior or posterior	M	0/32	0/29	1/29	2/34 (i)
	F	1/37	1/36	1/38	4/32 (i)
Chromodacryorrhea (secondary)	M	9/32	6/29	9/29	2/34 (d)
	F	20/37	21/36	14/38 (d)	2/32 (d)
Corneal dystrophy (focal scar)	M	16/32(50%)	16/29(55%)	22/29(76%) (i?)	31/34(91%) (i)
	F	23/37(62%)	29/36(81%) (i?)	29/38(76%) (i?)	31/32(97%) (i)

1. Not all signs present - diagnosis is based on the presence of choroidal pallor and the apparent absence of small choroidal vessels.
(i) considered to be increased due to fenthion treatment; (d) considered to be decreased due to fenthion treatment.

The above table shows that the high dose female group (100 ppm fenthion) has increased incidents of "diffuse posterior subcapsular cataract" and "possible retinal degeneration" (Dr. Rubin's report states that the cataract is a common sequel to retinal degeneration). The increases in these conditions is pronounced. There was also evidence of "phthisis bulbi" and "synechia" being elevated in females in the high dose group. These conditions were not considered truly compound related by the study report author and in some cases (including inflammation of the Harderian glands) were considered to be related to blood sampling from the retro-orbital bleeding.

The condition "chromodacryorrhea" was indicated as being decreased in the high dose male and mid and high dose female groups. The decrease in this condition may reflect an inability of the tear function of the eye to operate properly.

There appears to be a dose related occurrence of corneal dystrophy (focal scar), with an elevated incidence in all treated females and in levels 2 (20 ppm) and 3 (100 ppm) males. However the incidence is so high in all groups that the higher incidence probably may not be statistically or biologically significant for the groups other than the high dose group. Only the high dose group had accompanying signs of toxicity (particularly neovascularization and mineralization). Thus, only in the high dose group is this condition considered to be related to the test material.

2. Electroretinogram (ERG). Assessments were made on at least ten rats/sex/dose level during week 75 and prior to study termination. The test rats were given a limited ophthalmological examination which did not indicate compound related effects prior to the ERG. The rats were placed in an unlighted animal room the night before and during the exam. The selected rats were anesthetized with Halothane and electrodes were attached to the animal and a light flash was used to stimulate a retinal response. The response was measured and recorded on a floppy diskette. The results are summarized in Table 6 as follows:

Table 6. Results of Electroretinogram analysis of rats dosed with fenthion.

	Week 75 data				//	Prior to sacrifice data			
Acceptable ERGs:									
Males -	9,	7,	9	and 8	//	5,	8,	7	and 6.
Females -	8,	6,	2*	and 1*	//	8,	10,	7	and 0*.
ERGs not detected:									
Males -	0,	2,	0	and 0	//	2,	0,	0	and 1.
Females -	0,	1,	2	and 8*	//	1,	0,	0	and 10*.
ERGs suppressed in M-2:									
Males -	2,	1,	1	and 2	//	3,	2,	3	and 3.
Females -	2,	3,	6	and 1	//	1,	0,	3	and 0.
Total Males	11	10	10	10	//	10	10	10	10
Females	10	10	10	10	//	10	10	10	10

Data are number of animals with condition for the control, low, mid and high dose groups respectively.
* $p < 0.05$, study report statistics.

Table 6 shows that for females have there is clearly a decrease in acceptable ERGs in the mid and high dose groups at week 78 but only for the high dose group at termination. There is a corresponding increase in suppressed ERGs for the mid dose

group and increase in ERGs not detected for the high dose group at week 78. At termination none of the females had detectable ERGs in the high dose group and there was a slight (1 for the control vs 3 for the mid dose group) increase in suppressed ERGs

In summary, based on the decreases in acceptable and/or the presence of or suppressed ERGs, the NOEL/LEL was set at 5/20 ppm for females. At 100 ppm there is clearly an increase in ERGs not detected. There was no obvious effect in males (NOEL > 100 ppm).

3. Pathology/histology.

The eyes and optic nerve were removed from the carcasses and fixed in "universal fixative" or formalin depending on the mode of death. They were routinely stained with hematoxylin and eosin and examined. No special stains or histopathological techniques were utilized. Table 6 illustrates the microscopic findings in the eye and optic nerve in this study.

Table 7. Pathological findings in the eye as indicated by microscopy.

Lesion		Dose Level[1] Control	5 ppm	20 ppm	100 ppm
Eyes-Atrophy[3] (diffuse retinal) unilateral	M	6	6	5	7
	F	5	3	5	40*
bilateral	F	0	0	1	39
Eyes-retinal degeneration[3]	M	34 (68%) [1][2]	32 (64%)	25 (50%)	37 (74%)
	F	34 (68%)	34 (68%)	43 (86%)	1 (2%)
Eyes-Neovascularization corneal	M	4 (8%)	2 (4%)	4 (8%)	13* (26%)
	F	4 (8%)	3 (6%)	7 (14%)	29* (58%)
Optic nerve - Atrophy, unilateral	M	3/46 (6%) [1/19]	3/48 (6%)	6/49 (12%)	11/46*(23% [2/20]
	F	6/47 (12%) [0/20]	6/48 (12%)	3/47 (6%)	15/46*(32% [2/19]
Bilateral	F	0	0	0	5*
Corneal mineralization	M	4	2	4	13*
	F	4	3	7	29*

1. Incidents/50 animals for eyes. For optic nerve, the denominator indicates the number of animals examined.
2. Data in [] indicates observations at the interim sacrifice for 20 rats unless otherwise indicated by a denominator. If no entry there were none reported.
3. Atrophy is interpreted as an advanced stage of retinal degeneration, Thus, the noted decrease in degeneration is interpreted to be the increase in atrophy.
4. * $P < 0.05$. Study report statistics. Note data entered are from the text of the

study report and not from the pathology tables which report total mineralization for all eye structures.

The above table shows that the eyes have "atrophy" in the high dose group females. Males are not obviously affected. The report describes the atrophy in males and in the lower doses (< 20 ppm) for females as being primarily unilateral and being associated with the orbital bleeding techniques. A single 20 ppm female and nearly all of the 100 ppm females had bilateral atrophy more strongly suggesting a test chemical effect. The diffused retinal atrophy was described as involving the loss and/or thinning of the rod and cone layer, outer nuclear layer and outer plexiform layer. The loss of these layers was correlated with the ERG and ophthalmic results. Only the inner nuclear layer, inner plexiform layer and ganglion cell layer remained.

Most (68 to 86%) of the females have retinal degeneration in the control, low and mid dose group but there is only a single incident in the high dose group but the condition of the eyes in this group is not described. The female mid dose group (86%) appears to have more of this condition relative to the control and low dose groups (both 68%). The males are apparently not affected since the control (68%) is only slightly lower than the high dose group (74%).

Both the male high dose group and the female mid and high dose group have higher rates of neovascularization.

Optic nerve atrophy (unilateral) was increased in both males and females in the high dose groups and the male mid (not significant) dose group. The atrophy may be starting to become evident after 1 year. The study report asserts that this lesion is often associated with orbital eye bleeding. In females, however, the increase in bilateral optic nerve atrophy is more likely associated with the test material. In the absence of supporting data for other evidence of ocular effects in males, TB-I does not conclude that the apparent increase in atrophy in the eye is compound related. This is supported by the data with females which have the more apparent effects as indicated by ophthalmoscopy, ERG and histopathology. The females also have bilateral optic nerve atrophy.

In addition to the lesions mentioned above, the eyes of the female high dose group were noted to be the highest (10% affected vs 4% in the control) with acute inflammation and chronic inflammation was noted in the high dose group females (3 incidents or 6%) but only one other animal in all the other groups had this condition. It is likely that the inflammation was related to the other pathological conditions of the eye.

CONCLUSION (ophthalmoscopy, eye and optic nerve effects). NOEL/LEL = 5/20 ppm. At 20 ppm: ERGs suppressed (F); retinal degeneration (F, histologically); neovascularization (F). At 100 ppm: diffuse posterior subcapsular cataract (F) and retinal degeneration (F, ophthalmoscopy); atrophy (F, eye), neovascularization (M); optic nerve atrophy (F), retinal degeneration (M), retinal atrophy (F) and corneal mineralization (M/F); phthisis bulbi (F) and synechia (F).

9B. **Epididymides.** The epididymides was recognized as being affected by the author of this study report and the rat multi generation reproduction study in the Charles River CD strain rat (MRID No.:413486-01 and HED Document No.: 008545 dated Sep. 3, 1991) also indicated this structure to be affected by fenthion. In the reproduction study, the NOEL/LEL = 2/14 ppm and the condition was described as cytoplasmic vacuolation of epithelial ductal cells of the epididymides. At higher levels (100 ppm) epididymal weight was decreased.

The epididymides in this current study based on data in the Pathology Incidence Report (p. 437) for the <u>main phase</u> of this study had a condition described as "degeneration, vacuolar" but the background rate was too high to conclude that there was an effect of the test material. For example, there were 41/50(82%), 47/50(96%), 41/50(82%) and 48/50(96%) males affected for the control to high dose groups respectively.

The <u>interim sacrifice group</u> (page 573) was reported as having 0 males with epididymal vacuolization but <u>20/20</u> males in the high dose group had this condition. Note: The low and mid dosing levels were not included in the interim sacrifice group].

The <u>text</u> (data were not in the pathology tables) of the report presents additional findings which are summarized in Table 8 (lesion identification: followed by the incidents out of 50 males for the control to high dose groups respectively):

Table 8. Pathology of the epididymides in rats dosed with fenthion.

Proximal body vacuolar degeneration:	40, 46, 40, 7*.
Distal body vacuolar degeneration:	18, 16, 11, 0*.
Diffuse vacuolar degeneration of the epithelium of the body:	0, 0, 0, 35*.
Fine vacuolization in the epithelial cells in the head region:	0, 0 , 4, 43*.

Data are number of incidents for the control, low, mid and high doses respectively based on 50 animals per dose group.

CONCLUSION (pathology of the epididymides): NOEL/LEL = 5/20 ppm. At 100 ppm it is apparent that something is causing vacuolation changes in this structure. It also appears that 20 ppm may be a threshold dose.

9C. Nasolacrimal duct. This structure (listed in the pathology individual data sheets and summary tables under skull) was recognized as being affected by fenthion in the study report. The following number of male or female rats out of the 50 examined were reported as having the condition "vacuolar degeneration of the lacrimal duct":

males: 3, 2, 5 and 37*.

-females: 3, 7, 26* and 44*.

In addition, the following number of males or females were reported as having "foreign material in the nasal passages" out of 50 examined:

-males: 4, 6, 7 and 13.

-females: 2, 1, 3 and 11*.

CONCLUSION (nasolacrimal duct): NOEL/LEL = 5/20 ppm. 20 ppm: vacuolar degeneration in females. 100 ppm: in males.

Note: Due to the low level of response and because it was noted at the highest dose only, the foreign material in nasal passages is not considered of meaningful significance.

9D. Lung. Lung weight was decreased in the mid an high dose test groups for males. The study report indicated that the lung

was associated with higher incidence of "pneumonia primarily granulomatous with giant cell formation and bronchial epithelial hyperplasia". The following number of rats (out of 50 examined) were reported as having "pneumonia" for the control to high dose groups respectively:

-males: 8, 14, 16* and 22*
-females: 2, 0, 3 and 18*
* $p < .05$ study report statistics.

The report stated that this general type of pneumonia was similar to an "inhalation pneumonia".

CONCLUSION (lungs) : NOEL/LEL = 5/20 ppm. At 20 ppm pneumonia (M). At 100 ppm: pneumonia (M/F).

Note: In males, the low (5 ppm) dose group may be also have increased incidence of pneumonia although the increase is not statistically significant.

[The report presents an explanation for the "pneumonia" and possibly also the vacuolar degeneration in the nasolacrimal duct. This explanation provides that the increased nasal secretions resulting from inhibition oh ChE/AChE is responsible for the vacuolization and "pneumonia". The study did not however, clearly show that there were more animals having CHE/AChE symptoms to the extent that increased secretions especially in males were evident. There is also no correlation between there being more males affected with the "pneumonia but more females are affected with the nasolacrimal vacuolization. TB-I considers this a novel explanation but similar "pneumonia" and nasolacrimal vacuolization are not known (within the experience of TB-I reviewers) to be encountered in other studies with organophosphates and carbamates. The NOEL/LEL will still be included in the conclusions of the study.]

9E. Mineralization. The aorta, heart, lung, kidney, stomach, spinal cord, thyroid as well as others had either apparently compound related increases in mineralization or this condition was present in many of the animals. Table 7 illustrates the several organs which had apparently dose related increases in mineralization.

Mineralization is often associated with cell death but there was no increase in necrosis or other indication of cell death.

Table 9. Selected organs with compound related increases in mineralization, Dose Level[1]

Organ		Control	5 ppm	20 ppm	100 ppm
Stomach	M	1 [0][2]	2	5	32 [1]
	F	2	2	1	27
Heart	M	26 [6]	36	34	38 [11]
	F	16 [1]	32	25	32 [2]
Kidneys	M	30 [16]	40	42	47 [12]
	F	42 [16]	45	41	46 [18]
Spinal Cord	M	16	21	19	27
	F	8	13	1	15

1. Number of incidents based on 50 animals examined.
2. The data in [] is for the interim sacrifice of 20 rats/sex.

The data were not presented so as to indicate the degree of severity for mineralization so TB-I cannot determine if the degree of severity increased with the dose level. Mineralization in the stomach (the organ with the most obvious increase in incidence with dose level) was described as "raised zones" primarily seen in both 100 ppm males and females and appeared as focal to multifocal area of mineralization in the outermost muscular layer, muscularis externa". The interim sacrifice data do not indicate effects at one year for the stomach but may for the heart.

CONCLUSION (mineralization): NOEL/LEL = 20/100 ppm. 100 ppm: mineralization in the stomach. Other organs may also be affected but the high background rate for this condition in the controls obscures a more definite conclusion.

9F. Thyroids and parathyroids. The increase in mineralization may be related to a parathyroid effect which in turn affects the hormonal control of Ca++ metabolism. The study lists that 50 of each sex were examined for parathyroids and thyroids.

There were no compound related parathyroids lesions in males. There were a total of three adenomas, two in the controls and one in the high dose group. There were two incidents of hyperplasia in the control group. No other lesions were noted.

There were also very few females affected with para-

thyroid lesions. There were two adenomas, both in the mid dose group, one incident each of hyperplasia and "angiectasis" in a low and a mid dose group.

In the thyroids, among the females there was a decrease in the number of rats having c-cell hyperplasia with there being 18%, 14%, 26% but only 4% affected in the high dose group. This condition was evenly distributed among the male test groups. Both follicular and c-cell adenomas and carcinomas were present but there was no indication of a dose response.

CONCLUSION. There is no indication that pathological changes in the thyroid or parathyroid were responsible for the increased mineralization.

9G. Kidney. The mineralization may possibly relate to a pathological condition of the kidney resulting in impairment of the excretion of Ca++. Kidney weight was slightly decreased in the female high dose group. The report indicated that 50 kidneys of each sex per dose group were examined.

"Chronic nephropathy" was lowest in the high dose (44%/6%) when compared to the control (76%/16%) for both males/females. The low and mid dose groups had incidence frequencies similar to the controls. Other lesions did not show dose related increases in incidence.

CONCLUSION. There is no indication that the kidney was affected by the test material except for possible increases in mineralization in the high dose group (see Table 7). The apparent decrease in "chronic nephropathy" is considered a spurious finding.

9H. Skin. There were increased incidents of certain skin lesion in the high dose group for both males and females as follows (data are from the text of the report and were not traced to the pathology tables).

-on hindlimb	males:	5, 8, 9 and 26*
	females:	0, 0, 1 and 8*
-on tail	males:	14, 16, 22 and 43*
	females:	3, 4, 4 and 34*

These lesion were described as "a thickening of the squamous epithelium (acanthotic reactive hyperplasia) with elongated rete peg formation. On the surface, considerable keratin and debris often were noted. Epidermal inclusion, cysts, abscesses and ulceration also were occasionally present".

CONCLUSION (skin lesions): NOEL/LEL = 20/100 ppm. At 100 ppm there are lesions on the hindlimbs and tail. These may result

from irritability.

009870

9I. Harderian Gland. There was a higher rate of "Inflam./Chr.Act" in the high (32%/30%) as compared to the controls (16%/20%) for the males/females. Necrosis was also elevated in the high (26%), and mid (14%) dose females when compared to the control (10%) and low dose group (0%). These conditions are not considered primary or direct results of the test material. It may result from blood sampling from the orbital sinus.

9J. Larynx. There was a higher rate of "acute inflammation" in the high dose (26%) female group as compared to the control (12%). In general. other organs tended to have higher incidence of inflammation in the high dose groups also. The inflammation is noted but is not regarded as a specific response.

9K. Liver. Liver weight was decreased for the high dose males and females. The liver is a common target for chemical induced neoplasms. The liver has been implicated as a possible target organ for at least one other organophosphate (malathion). 50 of each sex/group were reportedly examined.

Liver hypertrophy was evident in only two mid dose group males and one low dose female. There were a total of 12 adenomas in the males: 6 in the controls and 3 each in the low and high dose group. There were a total of eight adenomas in the females: 3, 2, 2 and 1 for the control to high dose group respectively. There were four carcinomas among the males: one each in the control and mid dose group and 2 in the low dose group. There was a single carcinoma in the high dose group females.

9L. Brain. Brain weight was slightly elevated. A literature report (Veronesi et al, TAP 104:440-456, 1990 "The Neurotoxicity of Subchronic Acetylcholinesterase (AChE) Inhibition in the Rat Hippocampus", refer to HED Doc.: No.: 8309 for review) reported on a supposedly specific pattern of "gliosis, necrosis and cell dropout". The observations reported by Veronesi were not reported in this study.

9M. Testis. The high dose group had decreased weight but this was not statistically significant. The epididymides had vacuolation in a dose dependent manner. This condition may have resulted from a pathological condition of the testis.

Nearly all of the males for all dose groups had interstitial cell tumor. For example, there were 46, 47, 46, and 46 rats affected for the control to high dose groups respectively. A question is raised as to whether or not fenthion

affected the earlier onset of this tumor since there were no incidents in the control but two incidents in the high dose group at the 1 year interim sacrifice. Since 85% (18/21) of the controls and 84% (16/19) in the high dose group that died prior to termination had this tumor, the possibility of compound induced earlier onset for this tumor is considered unlikely by TB-I.

9N. Pituitary. Gross pathology indicated that "pituitary masses" were elevated in all treated females but there was no statistical significance and no compound relationship. Adenomas in this gland were common. There were 18, 17, 22 and 25 of 50 males affected and in addition 1 mid dose group male had a carcinoma. There were 23, 36, 28 and 26 females affected with adenomas. There were no carcinomas in the females.

10. DISCUSSION. Table 8 below compares the interpretation of the study data provided by the study report with TB interpretation.

Table 10. Comparison of the study report conclusions and TB-I conclusions and comments.

Study Report	TB-I Assessment/Comment
Clinical Signs: NOEL/LEL = 20/100 ppm. At 100 ppm: urine stain, alopecia, hunched back, loose stool, rough coat, eye opacity, lesions on tail and paws, enlarged preputial gland, and "irritation of the penis".	NOEL/LEL = 5/20 ppm. TB-I considers that a sufficient number of animals in the 20 ppm dose group were affected to judge that this level is the threshold level.
Mortality: NOEL > 100 ppm (HDT).	Concurs. Slight increase in high dose female deaths not supported by other signs of toxicity to conclude compound relationship.
Body Weight and Gain: NOEL/LEL = 20/100 ppm. Both males and females affected at 100 ppm.	Concurs. Weight affect is in adults.
Ophthalmoscopy: See special section.	Fenthion affects the eye and optic nerve. See below.
Hematology: NOEL > 100 ppm (HDT).	Concurs.

Clinical Chemistry: (Non ChE and AChE). NOEL > 100 ppm (HDT).	Concurs.
Urinalysis: NOEL > 100 ppm (HDT).	Concurs.
Organ Weight: NOEL > 100 ppm (HDT).	NOEL/LEL = 5/20 ppm. At 20 ppm: Lung weight decreased in males. Weight changes in the brain (slight increase), testis and liver are considered to be related to body weight change and were not supported by associated pathology,
Non-neoplastic Pathology: LELs: Pneumonia*: 20 ppm (M), 100 ppm (F). ..... Vacuolar degeneration of the naso-lacrimal duct*: 20 ppm (F), 100 ppm (M). ..... Vacuolar degeneration of the epididymides*: 100 ppm ..... Mineralization of the stomach musculature*: 100 ppm (M/F) ..... Chronic active dermatitis with crusts/debris, cysts and ulcers on feet and tails*: 100 ppm (M/F). *Said to be a secondary effect of ChE/AChE inhibition.	Concurs ..... Concurs ..... NOEL/LEL = 5/20 ppm. Vacuolation noted at 20 ppm and may be threshold. ..... Concurs ..... NOEL/LEL = 5/20 ppm. Threshold for effects considered to be 20 ppm. Study author's suggestion that the above pathological conditions are related to ChE/AChE inhibition is not supported by results with other organophosphates and carbamates (based on the experience of TB-I reviewers) but may be possible.
Neoplastic Pathology: "no overall significant increase in animals with tumors of either sex. No induced oncogenicity was established for fenthion in this study"	Concurs. No evidence of fenthion induced carcinogenicity.

Special Assessments: Eye and optic nerve effects. ..... Ophthalmoscopy: NOEL/LEL = 20/100 ppm. At 100 ppm: retinal degeneration, cataract formation and/or corneal scars. ..... ERGs: NOEL/LEL = 5/20 ppm for females. 100 ppm for males. At 20 and 100 ppm in females unacceptable and/or suppressed ERGs. No effects in males. ..... Histopathology: NOEL/LEL = 5/20 ppm for females and 20/100 ppm for males. At 5 ppm: bilateral retinal degeneration (F). At 100 ppm: neovascularization (M/F); mineralization of the cornea (F); bilateral optic nerve atrophy (F).	..... NOEL/LEL = 5/20 ppm. At 20 ppm: females have decrease in chromodacryorrhea. At 100 ppm males and females have decrease in chromodacryorrhea and retinal degeneration, cataract formation and/or corneal scars. [TB-I considers that the effect on chromodacyorrhea may indicate an adverse effect on the tear function of the eye.] ..... Concurs ..... Concurs. [Optic nerve atrophy was increased in both males and females but only in females was the condition bilateral. The males did not have other associated conditions to indicate that the increase in this condition was related to dosing.]
ChE/AChE: Plasma ChE: NOEL < 5 ppm, RBC AChE: NOEL/LEL = 5/20 ppm. Brain AChE: NOEL/LEL = 5/20 ppm.	NOEL < 5 ppm for plasma ChE, RBC and brain AChE. LEL = 5 ppm. Consistent inhibition of activity is noted at 5 ppm. Although it does not always exceed 20%, the consistency in the inhibition is considered more important.
Overall Conclusion for NOEL/LEL:	NOEL not established for this study based on ChE/AChE inhibition data.
Adequacy of dosing levels: No comments.	Dose levels are considered adequate for assessment of carcinogenicity.

CONCLUSION: This chronic feeding aspects of this study are classified as CORE MINIMUM[1]. The carcinogenicity aspects of this study are classified as CORE GUIDELINE. The following "one liner" is supported:

LEL (plasma ChE and RBC and brain AChE) = 5 ppm. NOEL not established. Moderate inhibition of plasma ChE (40%) and minimal inhibition of RBC (up to 19% and brain (14%) AChE at this level. Testing at lower levels not required at this time.

NOEL/LEL (Systemic effects): 5/20 ppm. At 20 ppm: epididymal pathology; vacuolation of nasolacrimal duct (F); pneumonia (M); lung weight (M); skin lesions and clinical signs (threshold, M/F). At 100 ppm: Body weight decreases in adults (M/F), mineralization (stomach and other structures); vacuolation of nasolacrimal duct (M).

NOEL/LEL (eye and optic nerve effects): 5/20 ppm. Eye function and/or pathology (F): At 100 ppm: Additional eye and optic nerve pathology (atrophy and/or neovascularization).

Carcinogenicity: No evidence of carcinogenicity.

Dose levels tested: 0, 5, 20 and 100 ppm corresponding to 0, 0.09, 0.3 and 1.8 mg/kg/day for males and 0.07, 0.3 and 1.5 mg/kg/day for females. Fischer 344 strain rat.

1. The study does not show a NOEL for ChE/AChE inhibition and precludes a classification as GUIDELINES.

Appendix I. Basis for Selection of the Dose Level for the Chronic Feeding/Carcinogenicity Study.

The dose levels were selected based on a 16 week feeding study in which the rats were dosed with 0, 2, 3, 5, 25 and 100 ppm. This specific study has not been reviewed by Toxicology Branch or its predecessor. The study was reported to establish a NOEL/LEL of 3/5 ppm based on ChE inhibition particularly in females. The rats were reported as tolerating the chemical up to 100 ppm but at this level the average serum and RBC inhibition was 70% and brain AChE was inhibited 50%.

Based on these observations, the choice of 5 ppm as the lowest test dose for the chronic feeding study is questionable since the earlier study already indicated inhibition at 5 ppm. The chronic feeding study eventually confirmed the effects of fenthion as an inhibitor of plasma CHE and RBC and brain ACHE.

Fenthion

Mobay Corporation

07-271-01

Dec. 17, 1990

MRID # 417431-01 (5 volumes)

009870

83-5 Chronic Feeding/Oncogenicity in the Rat

ACCEPTANCE CRITERIA

Does your study meet the following acceptance criteria?:

1. ✓ Technical form of the active ingredient tested.
2. ✓ At least 50 rats/sex/group (3 test groups and control group).
3. ✓ Dosing duration is at least 24 months.
4. ✓ Number of survivors in any group does not fall below 50% at 18 months or 25% at 24 months.
5. ‡ ? Doses tested include an MTD or limit dose if nontoxic (1000 mg/kg).
6. • No Doses tested include a NOEL.
7. • ✓ Analysis for test material stability, homogeneity and concentration in dosing medium
8. ___ Individual daily observations.
9. ___ Individual body weights.
10. ___ Individual or cage food consumption.
11. • ✓ Opthalmoscopic examination (at least pertest and at term) control and high dose.
12. ✓ Clinical pathology data for at least 10 rats/group consisting of 13, 14 & 15
13. ✓ Hematology at 6 month intervals consisting of at least;
    - ✓ Erythrocyte count
    - ✓ Hemoglobin
    - ✓ Hematocrit
    - ✓ Leucocyte count
    - • ✓ Differential count
    - ✓ Platelet count (or clotting measure)
14. ✓ Clinical chemistry at 6 month intervals consisting of at least;
    - ✓ Alkaline phosphatase
    - ✓ Aspartate aminotransferase
    - • ✓ Creatinine kinase
    - ✓ Lactic dehydrogenase
    - ✓ Glucose
    - ✓ Bilirubin
    - ✓ Cholesterol
    - • ✓ Creatinine
    - ✓ Total Protein
    - ✓ Albumin
    - ✓ Urea (as BUN)
    - ✓ Inorganic phosphate
    - ✓ Calcium
    - • ✓ Potassium
    - ✓ Sodium
    - • ✓ Chloride
15. ✓ Urinalysis at 6 month intervals consisting of at least;
    - ✓ Blood
    - ✓ Protein
    - ✓ Ketone bodies
    - ✓ Appearance
    - ✓ Glucose
    - ✓ Total bilirubin
    - • ✓ Urobilirubin
    - ✓ Sediment
    - ✓ Specific gravity (osmolality)
    - • ___ Volume
16. ___ Individual necropsy of all animals.
17. ___ Histopathology of the following tissues performed on all nonrodents and rodents, all control and high dose animals, all animals that died or were killed on study, all gross lesions on all animals, target organs on all animals and lungs, liver and kidneys on all other animals.
    - ✓ aorta
    - ✓ jejunum
    - ✓ peripheral nerve

Criteria marked with a • are supplemental and may not be required for every study.

✓ eyes	✓ bone marrow	✓ kidneys†
✓ caecum	✓ liver†	✓ esophagus
✓ colon	✓ lung†	✓ ovaries†
✓ duodenum	✓ lymph nodes	✓ oviduct
✓ brain†	✓ stomach	✓ pancreas
✓ skin	✓ mammary gland	✓ rectum
✓ heart†	✓ spleen†	✓ spinal cord (3x)
✓ testes†	✓ musculature	✓ thyroid / parathyroids
✓ pituitary	✓ epididymis	✓ salivary glands
✓ ileum	✓ adrenals†	✓ thymus
✓ trachea	✓ uterus	✓ urinary bladder

† organs to be weighed.

‡ The position document entitled "Selection of a Maximum Tolerated Dose (MTD) in Oncogenicity Studies (EPA No. 540/09-88-005) stated EPA's criteria for determining if an oncogenicity study has been adequately performed in terms of doses tested. However OPP is also aware that older oncogenicity studies, upon initial review or re-review, may have been tested at doses lower than the predicted MTD. In the event that such testing appears to be at doses less than the predicted MTD, the Office of Pesticides Program has been reviewing and considering the entire weight of the evidence to determine if retesting is necessary. Certain factors which affect the agency's decision to retest include but are not limited to the following: demonstrated oncogenicity in another species, nearness to the apparent MTD, genotoxic effects, structure-activity factors, absolute value of the highest dose tested and metabolic considerations.

Criteria marked with a * are supplemental and may not be required for every study.