(3)

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

OFFICE OF
PREVENTION, PESTICIDES, AND
TOXIC SUBSTANCES

MEMORANDUM

June 18, 2004

Subject: EPA Id No.: 083601. Triphenyltin hydroxide. Review of a subchronic neurotoxicity study (2001, MRID No.: 45546001).

TXR # 0050309
DP Barcode No.: D279514
Submission No.: Not provided
PC Code: 083601

From: John Doherty
ReRegistration Branch III
Health Effects Division 7509C

To: Robert McNally
and
Wilhelmena Livingston
Risk Manager #60
Special Review and ReRegistration Division 7507C

Through: Catherine Eiden
Branch Chief
ReRegistration Branch III
Health Effects Division 7509C

Background, Conclusions and Comments.

ReRegistration Branch III has reviewed the subchronic neurotoxicity study (2001, MRID No.: 45546001) with triphenyltin hydroxide and determined that the study is ACCEPTABLE/Guideline and satisfies the requirement for a series 870.6200 subchronic neurotoxicity study in rats. A copy of the DER is attached and the study is further identified and the NOAEL and LOAEL provided in the following table.

AUG 24 2004

Table. Study Reviewed.

Study Identification	Executive Summary
870.6200. Subchronic Neurotoxicity - rats. WIL Research Laboratories, Study No.: WIL-399003, November 12, 2001.	In a subchronic neurotoxicity study (2001, MRID 45546001), fentin hydroxide (96.0% a.i., Lot # ZVRAM.928K) was administered in the diet for 13 weeks to 10 Crl:CD® (SD)IGS BR rats/sex/group at doses of 0, 5, 20, or 80 ppm (equivalent to 0, 0.4/0.4, 1.4/1.6, and 5.7/6.1 mg/kg bw/day in males/females). Neurobehavioral assessment (functional observational battery and motor activity testing) was performed in 10 animals/sex/group prior to treatment, and at Weeks 3, 7, and 12. At study termination, 5 animals/sex from the control and 80 ppm groups were euthanized and perfused *in situ* and subjected to histopathological evaluation of brain and peripheral nervous system tissues. At 80 ppm, body weight was decreased throughout the study in males, and the decrease was significant ($p<=0.05$) at Weeks 3-6 (decr 11-12%). Body weight gains at Weeks 0 to 1, 1 to 2, and 2 to 3 were decreased ($p<=0.05$) by 21-39%, and overall (Weeks 0-13) body weight gain was decreased (not statistically significant [NS]) by 12%. In females, body weight was consistently decreased (NS) by 3-6% from Week 2 until the end of the study. Body weight gain was generally decreased (NS) in females by 8-100% throughout the study, and was significant ($p<=0.01$) at Week 1 to 2 (decr 53%). Food consumption was decreased ($p<=0.05$) at Weeks 0 to 1 and 2 to 4 by 12-13% in the males and at Weeks 0 to 6, 7 to 8, and 9 to 11 by 10-16% in females. Total motor activity was decreased throughout treatment in the males (decr 36-40%; $p<=0.05$) and in the females (decr 10-37%; NS). Additionally, ambulatory activity was decreased (NS) throughout treatment in the males (decr 27-40%) and females (decr 10-37%). Habituation was unaffected by treatment. No treatment-related effects were observed on mortality, clinical signs, brain weight, length, or width, or neuropathology. FOB testing revealed no treatment-related effects. **The LOAEL is 80 ppm (equivalent to 5.7/6.1 mg/kg/day in males/females), based on decreased body weight, body weight gains, food consumption, and total and ambulatory motor activity in both sexes. The NOAEL is 20 ppm (equivalent to 1.4/1.6 mg/kg/day in males/females).** The study is classified as **acceptable/guideline** and satisfies the guideline requirements (870.6200b) for a subchronic neurotoxicity study in the rat.

DATA EVALUATION RECORD

FENTIN HYDROXIDE (TPTH)

Study Type: §82-7a, Subchronic Neurotoxicity Screening Battery in Rats

Work Assignment No. 01-01-23 B (MRID 45546001)

Prepared for
Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by
Pesticides Health Effects Group
Sciences Division
Dynamac Corporation
2275 Research Boulevard
Rockville, MD 20850-3268

Primary Reviewer:
Ronnie J. Bever Jr., Ph.D.

Signature: Ronnie J. Bever Jr.
Date: 4/28/04

Secondary Reviewer:
David A. McEwen, B.S.

Signature: David A. McEwen
Date: 4/30/04

Project Manager:
Mary L. Menetrez, Ph.D.

Signature: Mary L. Menetrez
Date: 4/30/04

Quality Assurance:
Steven Brecher, Ph.D.

Signature: Steven Brecher
Date: 4/28/04

Disclaimer

This Data Evaluation Record may have been altered by the Health Effects Division subsequent to signing by Dynamac Corporation personnel.

AUG 24 2004 41.5

EPA Reviewer: John Doherty **Signature:** John Doherty
Reregistration Branch 3, Health Effects Division (7509C) **Date** June 3, 2004
Work Assignment Manager: Ghazi Dannan, Ph.D. **Signature:** Ghazi A. Dannan
Registration Action Branch 3, Health Effects Division (7509C) **Date** 6/8/04
Template version 11/01

DATA EVALUATION RECORD

STUDY TYPE: Subchronic Neurotoxicity- Feeding Study in Rats, OPPTS 870.6200b [§82-7]; No OECD guideline.

PC CODE: 083601 **DP BARCODE:** D279514
TXR#: 0050309 **SUBMISSION NO.:** None

TEST MATERIAL (PURITY): Fentin Hydroxide (TPTH; 96.0% a.i.)

SYNONYMS: Triphenyltin hydroxide

CITATION: Nemec, M.D. (2001) A dietary subchronic (90-day) neurotoxicity study of TPTH in rats. WIL Research Laboratories, Inc., Ashland, OH. Laboratory Study No.: WIL-399003, November 12, 2001. MRID 45546001. Unpublished.

SPONSOR: TPTH Task Force c/o Landis International, 3185 Madison Highway, Valdosta, GA

EXECUTIVE SUMMARY - In a subchronic neurotoxicity study (2001, MRID 45546001), fentin hydroxide (96.0% a.i., Lot # ZVRAM.928K) was administered in the diet for 13 weeks to 10 Crl:CD® (SD)IGS BR rats/sex/group at doses of 0, 5, 20, or 80 ppm (equivalent to 0, 0.4/0.4, 1.4/1.6, and 5.7/6.1 mg/kg bw/day in males/females). Neurobehavioral assessment (functional observational battery and motor activity testing) was performed in 10 animals/sex/group prior to treatment, and at Weeks 3, 7, and 12. At study termination, 5 animals/sex from the control and 80 ppm groups were euthanized and perfused *in situ* and subjected to histopathological evaluation of brain and peripheral nervous system tissues.

At 80 ppm, body weight was decreased throughout the study in males, and the decrease was significant ($p<=0.05$) at Weeks 3-6 (decr 11-12%). Body weight gains at Weeks 0 to 1, 1 to 2, and 2 to 3 were decreased ($p<=0.05$) by 21-39%, and overall (Weeks 0-13) body weight gain was decreased (not statistically significant [NS]) by 12%. In females, body weight was consistently decreased (NS) by 3-6% from Week 2 until the end of the study. Body weight gain was generally decreased (NS) in females by 8-100% throughout the study, and was significant ($p<=0.01$) at Week 1 to 2 (decr 53%). Food consumption was decreased ($p<=0.05$) at Weeks 0 to 1 and 2 to 4 by 12-13% in the males and at Weeks 0 to 6, 7 to 8, and 9 to 11 by 10-16% in females. Total motor activity was decreased throughout treatment in the males (decr 36-40%; $p<=0.05$) and in the females (decr 10-37%; NS). Additionally, ambulatory activity was decreased (NS) throughout treatment in the males (decr 27-40%) and females (decr 10-37%). Habituation was unaffected by treatment. No treatment-related effects were observed on mortality, clinical signs,

brain weight, length, or width, or neuropathology. FOB testing revealed no treatment-related effects. **The LOAEL is 80 ppm (equivalent to 5.7/6.1 mg/kg/day in males/females), based on decreased body weight, body weight gains, food consumption, and total and ambulatory motor activity in both sexes. The NOAEL is 20 ppm (equivalent to 1.4/1.6 mg/kg/day in males/females).**

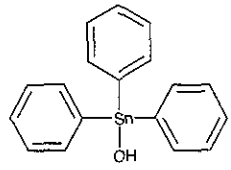
The study is classified as **acceptable/guideline** and satisfies the guideline requirements (870.6200b) for a subchronic neurotoxicity study in the rat.

COMPLIANCE - Signed and dated GLP, Quality Assurance, Data Confidentiality, and Flagging statements were provided.

I. MATERIALS AND METHODS

A. MATERIALS

1. **Test material:** TPTH (triphenyltin hydroxide)

Description:	Fine, white powder
Lot #:	ZVRAM.928K
Purity (w/w):	96.0% a.i.
Stability of compound:	Stable in the diet after storage at room temperature for 4 days
CAS #:	76-87-9
Structure:	

2. **Vehicle** - Diet

3. **Test animals**

Species:	Rat
Strain:	Crl:CD® (SD)IGS BR
Age and weight at Day 0:	44 days old; 167-232 g males; 132-177 g females
Source:	Charles River Laboratories (Raleigh, NC)
Housing:	Individually in suspended, stainless steel, wire-mesh cages
Diet:	Certified Rodent Lab Diet® #5002 (PMI Nutrition International, Inc.), *ad libitum*, except during neurobehavioral assessments
Water:	Reverse-osmosis purified tap water, *ad libitum*
Environmental conditions	
Temperature:	21.6-22.9°C
Humidity:	38.3-60.8%
Air changes:	Not reported
Photoperiod:	12 hrs light/12 hrs dark
Acclimation period:	13 days

B. STUDY DESIGN

1. **In life dates** - Not provided; approximately 1/25/01 to 4/19/01, based on dates given for the preparation of the diet

2. **Animal assignment and treatment** - Animals were randomly assigned to dose groups using procedure which insured homogeneity of group means and variances for body weights, as detailed in Table 1. The Sponsor stated that doses were selected based on the results of previous toxicity studies; however, data were not presented. Dietary formulations were removed daily from frozen storage and offered to the animals for 91 days.

Table 1. Study design [a]

Experimental Parameter	Dose (ppm)			
	0	5	20	80
Achieved chemical intake (mg/kg/day [M/F])	0/0	0.4/0.4	1.4/1.6	5.7/6.1
Number of animals	10/sex	10/sex	10/sex	10/sex
Behavioral testing (FOB, motor activity)	10/sex	10/sex	10/sex	10/sex
Neuropathology [b]	10/sex	10/sex	10/sex	10/sex

a Data were obtained from pages 22-26, 67, and 70 in MRID 45546001.

b Although tissues were perfused for 10 rats/sex/dose, only five randomly selected control rats/sex and five 80 ppm rats/sex were examined.

3. Test substance preparation and analysis - Dietary formulations were prepared weekly by adding the appropriate amounts of test substance to a small amount of diet to form a premix. The premixes were further diluted with the required amount of diet to achieve the test doses. Dietary formulations were stored frozen. Homogeneity (top, middle, bottom) and stability were tested on each dietary formulation on two occasions prior to dosing. Stability was evaluated twice for 4 and 8 days at room temperature and once for 15 and 22 days at room temperature and for 4 and 8 days frozen. Concentration of test material in the diet was determined in all dietary formulations prior to treatment, and at Weeks 1, 6, and 12.

Results

Homogeneity Analysis (range as % relative standard deviation): 3.1-8.0%

Stability Analysis (range as % of Day 0):

Days on Study	Dose (ppm)		
	5	20	80
Room Temperature			
4	66.1-100	76.3-98.3	79.5-87.3
8	45.9-101	73.1-93.1	94.5-95.3
15	81.8	70.2	87.4
22	49.6	53.6	60.1
Frozen			
4	84.0	91.7	86.0
8	76.5	95.3	131

Concentration Analysis (range of replicate means):

Dose (ppm)	% of Nominal
5	86.3-110
20	91.7-97.6
80	91.3-113

Considerable variation was noted between the two days of room temperature stability measurements of the 5 ppm dietary formulation. The Sponsor stated that the compound may bind to the diet, thus interfering with accurate analysis. According to the methodology, the diets were prepared weekly and stored frozen and aliquots were at room temperature for only 24 hours while being offered to the animals. The analytical data indicated that the mixing procedure was adequate and that the variation between nominal and actual dosage to the study animals was acceptable.

4. **Statistics** - Statistical significance of the data was tested using two-tailed analyses at $p \leq 0.05$ and 0.01. The data were analyzed using the following statistical methods:

Parameter	Statistical Method
Body weight, body weight gain, food consumption, brain weight data, continuous FOB data	One-way ANOVA, Dunnett's test
Discontinuous FOB data	Fisher's exact test
Locomotor activity	Two-way repeated measures ANOVA, one-way ANOVA, Dunnett's test

C. METHODS/OBSERVATIONS

1. **Mortality and clinical observations** - All animals were observed twice daily for mortality and moribundity. Detailed clinical observations were performed daily.

2. **Body weight** - All animals were weighed prior to initiation of treatment, weekly thereafter, and at termination. Additionally, all animals were weighed as part of the FOB.

3. **Food consumption** - Food consumption was measured daily, beginning one week prior to treatment. Weekly food consumption (g/animal/day) was reported. Mean daily intake of test material (mg/kg/day) was calculated for each sex and dose group from mean body weight and food consumption data.

4. **Neurobehavioral assessment**

a. **Functional Observational Battery (FOB)** - The FOB was performed on 10 animals/sex/dose prior to treatment, and at Weeks 3, 7, and 12. The scoring criteria and equipment description for the FOB were included in the Study Report, pages 935-950. Testing was performed by the same technicians when possible, and the technicians were blind to the animal group assignment. Home cage observations were performed in the animal room, and all other observations were

performed in a sound-attenuated room with 70±10 dB of white noise. Open field observations were performed during a 2 minute observation period.

The following CHECKED (X) parameters were examined:

	HOME CAGE OBSERVATIONS		HANDLING OBSERVATIONS		OPEN FIELD OBSERVATIONS
X	Posture*		Reactivity*	X	Mobility
X	Biting	X	Ease of removal from cage	X	Rearing+
X	Convulsions*	X	Lacrimation* / chromodacryorrhea	X	Arousal/ general activity level*
X	Tremors*	X	Salivation*	X	Convulsions*
	Abnormal movements*	X	Piloerection*	X	Tremors*
X	Palpebral closure*	X	Fur appearance		Abnormal movements*
	Gait abnormalities	X	Palpebral closure*	X	Urination / defecation*
X	Feces consistency	X	Respiratory rate+	X	Grooming
	SENSORY OBSERVATIONS	X	Red/crusty deposits*	X	Gait abnormalities / posture*
X	Approach response+	X	Eye prominence / pupil size*	X	Gait score*
X	Touch response+	X	Muscle tone*	X	Bizarre / stereotypic behavior*
X	Startle response*		Vocalizations	X	Backing
X	Pain response*	X	Skin color/mucous membranes/eye	X	Time to first step
X	Pupil response*	X	Ease of handling animal in hand		
X	Eyeblink response		**PHYSIOLOGICAL OBSERVATIONS**		**NEUROMUSCULAR OBSERVATIONS**
X	Forelimb extension	X	Body weight*	X	Hindlimb extensor strength
X	Hindlimb extension	X	Body temperature+	X	Forelimb grip strength*
X	Righting reflex+	X	Catalepsy	X	Hindlimb grip strength*
	Visual placing response			X	Landing foot splay*
	Pinna reflex		**OTHER OBSERVATIONS**	X	Rotarod performance
X	Olfactory orientation				

* Required parameters; +Recommended parameters

b. <u>Locomotor activity</u> - Following the FOB, each animal was individually placed in a clear plastic rectangular cage and movement was monitored for 60 minutes by a series of photobeams using the SDI Photobeam Activity System (San Diego, CA). Motor activity was measured during twelve 5-minute intervals. Ambulatory motor activity (the interruption of two or more consecutive photobeams) and total motor activity (the interruption of one photobeam) were quantified.

5. <u>Sacrifice and pathology</u> - At Week 13, all 10 animals/sex/dose were sacrificed by intraperitoneal injection of sodium pentobarbital, and the tissues were perfused *in situ*. Fixed brain weight (excluding olfactory bulbs) and brain dimensions (width as the widest part of the cerebral hemispheres and length at the rostral part of the cerebral hemispheres to the most caudal part of the cerebellum) were determined. The following CHECKED (X) tissues were collected.

	CENTRAL NERVOUS SYSTEM		PERIPHERAL NERVOUS SYSTEM
	BRAIN		**SCIATIC NERVE**
X	Forebrain		Mid-thigh
X	Center of cerebrum	X	Sciatic Notch
X	Midbrain		
X	Cerebellum		**OTHER**
X	Pons	X	Sural Nerve
X	Medulla oblongata	X	Tibial Nerve
	SPINAL CORD	X	Peroneal Nerve
X	Cervical swelling	X	Lumbar dorsal root ganglion
X	Lumbar swelling	X	Lumbar dorsal root fibers
	Thoracic swelling	X	Lumbar ventral root fibers
	OTHER	X	Cervical dorsal root ganglion
X	Gasserian Ganglion	X	Cervical dorsal root fibers
X	Trigeminal nerves	X	Cervical ventral root fibers
X	Optic nerve		
X	Eyes		
X	Gastrocnemius muscle		

Randomly selected samples from 5 rats/sex of the control and 80 ppm groups were collected, processed (embedded in paraffin or plastic, sectioned, and stained with hemotoxylin and eosin) and examined histologically.

9. **Positive controls** - Summaries of four studies (WIL-99032, WIL-99034, WIL-99035, WIL-99140, and WIL-99149) performed to generate positive control data and validate the procedures and observers of the performing lab to conduct the FOB and to assess motor activity, neurotoxicity were provided. Exposure to **3,3-Iminodipropionitrile** (IDPN, 2000 mg/kg, single gavage dose) induced the following in both sexes: (i) decreased body weight; (ii) FOB effects (eg. decreased mobility, impaired gait, retropulsion, and circling); (iii) decreased fore- and hindlimb grip strength in males; (iv) decreased startle response; and (v) impaired air righting reflex. **Acrylamide** (10 or 20 mg/kg, daily gavage doses 5 days/week for 4 weeks) induced the following effects in both sexes during the FOB at 14, 21, and 28 days after initial dosing: (i) alterations in muscle tone; (ii) alterations in startle and tail pinch responses and air righting reflex; (iii) decreased hindlimb extensor strength and fore- and hindlimb grip strength; (iv) reduced rotarod performance; (v) increased hindlimb foot splay; and (vi) decreased body weight and body temperature. Additionally, the following histopathological lesions were noted at 20 mg/kg: axonal degeneration, digestion chambers, swollen axon cylinders or demyelination in the trigeminal nerve, lumbar dorsal and ventral root fibers, cervical dorsal root fibers, sciatic, sural, tibial and peroneal nerves, lumbar root (females only), and cervical ventral root fibers (females only). **Trimethyltin chloride** (7.5 mg/kg, single i.p. dose) induced neuronal loss in the dentate gyrus and chromatolysis in the gasserian ganglion neurons in the males. **Carbaryl** (10 or 50 mg/kg, single i.p. dose) induced the following FOB effects: (i) altered posture, palpebral closure, and convulsions/tremors during home cage observations; (ii) altered ease of removal and handling, salivation, fur appearance, and eye prominence during handling; (iii) increased time to first step, alterations in mobility, gait, gait score, arousal, convulsions/tremors, and number of rears during open-field observations; (iv) alterations in approach, touch, startle, tail pinch, and

pupil responses, olfactory orientation, and air righting reflex; (v) alterations in hindlimb extensor strength, grip strength, and rotarod performance; and (vi) alterations in catalepsy time and body temperature. Additionally, total and locomotor activity were decreased. Total and locomotor activity were increased with **d-Amphetamine sulfate** (2 or 4 mg/kg, single i.p. dose) and decreased with **Chlorpromazine hydrochloride** (5 or 10 mg/kg, single i.p. dose). Inter-observer reliability was demonstrated using **3,3-Iminodipropionitrile** (IDPN, 2000 mg/kg, single gavage dose). All observers detected the FOB effects from IDPN (decreased mobility, impaired gait, retropulsion, circling, decreased fore- and hindlimb grip strength in males, decreased startle response, and impaired air righting reflex). In addition, a study was conducted to insure that the untreated rats approached normal activity levels by the last 20% of the session, in accordance to regulatory guidelines. In this study, juvenile Crl:CD®(SD)IGS BR rats were acclimated and maintained as in the subchronic neurotoxicity study. Each animal was individually placed in a clear plastic rectangular cage and movement was monitored for 120 minutes by a series of photobeams using the SDI Photobeam Activity System (San Diego, CA) with 70 dB background noise. Motor activity was measured during 1-minute intervals. The results of this test suggested the optimal duration for the locomotor activity test and the appropriate epoch within the test session.

II. RESULTS

A. OBSERVATIONS

1. Clinical signs - No adverse treatment-related clinical signs were observed. Decreased defecation and urination were observed in the ≥20 ppm females (3-5 treated vs 0 controls; 5-24 occurrences; Table 2). However, these findings occurred infrequently and were transient. They amy also relate to the decrease in food consumption also seen in this group. No other treatment-related clinical signs were observed.

Table 2. Selected clinical observations (total occurrence/number of animals) in female rats treated with TPTH for 13 weeks. [a]

Parameter	Dose (ppm)			
	0	5	20	80
Normal	636/10	717/9	620/10	722/10
Decreased defecation	0/0	1/1	5/3	24/5
Decrease urination	0/0	1/1	5/3	9/4

a Data (n=10) were obtained from pages 45-46 of MRID 45546001.

2. Mortality - All animals survived to scheduled sacrifice.

B. BODY WEIGHT AND BODY WEIGHT GAIN - In the 80 ppm males, body weight was decreased throughout the study, and the decrease was significant ($p \leq 0.05$) at Weeks 3-6 (↓11-12%; Table 3). Body weight gains at Weeks 0 to 1, 1 to 2, and 2 to 3 were decreased ($p \leq 0.05$) by 21-39%, and overall (Weeks 0-13) body weight gain was decreased (not statistically significant [NS]) by 12%. In the 80 ppm females, body weight was consistently decreased (NS) by 3-6% from Week 2 until the end of the study. Body weight gain was generally decreased (NS) by 8-100% throughout the study in the 80 ppm females, but was significant ($p \leq 0.01$) only at Week 1 to 2 (↓53%). The body weight and body weight gain of the 5 and 20 ppm dose groups were similar to the controls.

Table 3. Mean (±SD) body weights (g) at selected intervals in rats treated with TPTH in the diet for 13 weeks. [a]

Weeks on study	Dose (ppm)			
	0	5	20	80
Males				
0	205±16.4	202±17.8	197±18.1	200±18.9
3	333±26.2	334±31.7	333±30.9	294±27.9* (↓12)
6	389±36.6	397±43.9	394±38.8	347±30.5* (↓11)
13	489±50.6	506±51.2	506±54.1	451±38.8
BWG (0 to 1)	51±5.4	54±13.8	58±9.4	40±8.7* (↓22)
BWG (1 to 2)	39±4.7	40±6.4	40±8.1	31±7.4* (↓21)
BWG (2 to 3)	38±7.5	38±7.8	38±8.7	23±6.5** (↓39)
BWG (0 to 13)	284±40.2	304±41.2	309±39.6	251±24.9 (↓12)
Females				
0	152±9.3	158±9.1	158±9.9	156±11.1
2	195±14.3	203±14.2	203±15.7	187±15.4 (↓4)
9	264±21.4	277±22.9	277±26.3	249±31.0 (↓6)
10	266±21.9	283±22.1	282±18.9	258±25.6 (↓3)
13	287±26.3	301±22.6	300±21.4	273±22.2
BWG (0 to 1)	26±8.1	26±5.2	32±5.7	24±4.0 (↓8)
BWG (1 to 2)	17±3.1	19±5.3	13±6.2	8±7.3** (↓53)
BWG (8 to 9)	7±4.6	6±5.7	3±21.0	0±21.5 (↓100)
BWG (0 to 13)	135±22.2	143±15.0	142±16.1	118±13.8 (↓13)

a Data (n=10) were obtained from pages 47-58 of MRID 45546001. Percent difference from controls, calculated by reviewers, is included in parentheses.

* Statistically different ($p \leq 0.05$) from the controls

** Statistically different ($p \leq 0.01$) from the controls

C. FOOD CONSUMPTION AND MEAN CHEMICAL INTAKE - At 80 ppm, food consumption was decreased ($p \leq 0.05$) at Weeks 0 to 1 and 2 to 4 by 12-13% in the males and at Weeks 0 to 6, 7 to 8, and 9 to 11 by 10-16% in females (Table 4). Food consumption in the 5 and 20 ppm groups was similar to controls. Mean chemical intake (mg/kg/day) is presented in Table 1.

Table 4. Mean (±SD) food consumption (g/animal/day) at selected intervals in rats treated with TPTH in the diet for 13 weeks. [a]

Week	Dose (ppm)			
	0	5	20	80
Males				
0 to 1	24±16	24±1.4	25±2.3	21±3.1* (↓13)
3 to 4	25±1.6	25±2.5	27±2.3	22±1.9** (↓12)
Females				
0 to 1	18±1.5	18±1.1	18±1.6	16±1.5** (↓11)
2 to 3	19±2.0	20±1.6	19±1.5	16±1.4** (↓16)
10 to 11	20±1.6	20±1.8	20±2.7	18±1.8* (↓10)

a Data (n=10) were obtained from pages 59-64 of MRID 45546001. Percent difference from controls, calculated by reviewers, is included in parentheses.
* Statistically different ($p \leq 0.05$) from the controls
** Statistically different ($p \leq 0.01$) from the controls

D. NEUROBEHAVIORAL RESULTS

1. FOB findings - No treatment-related FOB findings were observed at any dose in either sex throughout the study. All differences were transient, unrelated to dose, and/or within the biological variation of the controls or pretest values.

2. Locomotor activity - At 80 ppm, total motor activity was decreased throughout treatment in the males (↓36-40%; $p \leq 0.05$) and in the females (↓10-37%; NS; Table 5a). Additionally, ambulatory activity was decreased (NS) throughout treatment in the 80 ppm males (↓27-40%) and females (↓10-37%; Table 5b). Habituation was unaffected by treatment. Total motor and ambulatory activity were similar to controls in the 5 and 20 ppm groups. Subsession data for the total motor and ambulatory activity counts are provided in Tables 5c through 5f.

Table 5a. Motor activity (total activity counts for session; mean±SD)[a]

Test Day	Dose (ppm)			
	0	5	20	80
Males				
Pre-test	541±198.8	489±220.6	559±258.1	496±220.3 (↓8)
Week 3	1037±365.8	995±346.7	978±230.3	663±160.0* (↓36)
Week 7	873±226.9	811±214.5	912±353.3	527±309.2* (↓40)
Week 12	879±232.6	843±281.9	795±274.7	566±210.5* (↓36)
Females				
Pre-test	649±444.6	477±123.3	614±240.2	481±286.1 (↓23)
Week 3	1078±476.4	975±438.9	1121±306.2	830±346.9 (↓10)
Week 7	969±488.1	805±368.2	996±284.2	707±193.5 (↓22)
Week 12	1161±480.1	941±331.4	955±290.9	761±286.1 (↓37)

a Data (n=10) were obtained from pages 223-224 of MRID 45546001. Percent difference from controls, calculated by reviewers, is included in parentheses.

* Statistically different ($p \leq 0.05$) from the controls

** Statistically different ($p \leq 0.01$) from the controls

Table 5b. Motor activity (ambulatory activity counts for session)[a]

Test Day	Dose (ppm)			
	0	5	20	80
Males				
Pre-test	160±63.9	137±68.1	170±94.8	145±89.7(↓9%)
Week 3	291±122.0	268±125.9	288±91.4	211±71.8(↓27%)
Week 7	214±57.7	205±78.7	236±115.3	129±83.2(↓40%)
Week 12	203±56.3	182±70.7	186±63.8	147±72.7(↓28%)
Females				
Pre-test	203±159.1	149±46.4	194±97.8	156±105.0(↓23%)
Week 3	343±170.8	351±190.5	395±112.4	307±132.8(↓10%)
Week 7	278±171.8	254±115.3	336±115.8	218±71.4(↓22%)
Week 12	395±225	297±121.8	313±95.1	247±113.3(↓37%)

a Data (n=10) were obtained from pages 223-224 of MRID 45546001.

Table 5c. Total activity subsession counts (mean±SD) in males [a]

Interval (min)	Dose (ppm)			
	0	5	20	80
Pretest				
0-15	389±151.6	298±99.6	421±173.4	332±141.9
16-30	84±96.7	85±78.0	64±63.8	72±39.7
31-45	54±149.9	52±106.9	46±79.9	27±49.9
46-60	14±20.7	54±106.2	28±35.6	66±133.5
Week 3				
0-15	663±137.2	602±197.8	613±179.4	486±152.7
16-30	209±155.7	225±129.0	207±124.1	129±132.6
31-45	101±181.9	137±147.4	126±134.2	37±44.7
46-60	65±118.4	32±47.6	32±60.1	11±21.0
Week 7				
0-15	507±154.9	523±175.8	482±193.6	336±128.9
16-30	169±122.1	182±94.9	207±71.4	51±60.1
31-45	151±110.7	60±80.4	146±139.9	39±69.7
46-60	47±66.0	47±71.6	78±124.6	100±158.5
Week 12				
0-15	575±139.6	557±145.1	568±115.6	399±181.9
16-30	155±79.6	157±140.7	129±103.4	101±72.4
31-45	111±135.6	88±90.4	60±73.9	31±40.8
46-60	39±62.7	41±48.3	39±43.7	36±63.1

a Data (n=10) were obtained from pages 648-679 of MRID 45546001.

Table 5d. Total activity subsession counts (mean±SD) in females [a]

Interval (min)	Dose (ppm)			
	0	5	20	80
Pretest				
0-15	422±184.0	391±90.0	376±198.8	388±164.1
16-30	141±193.7	23±19.3	52±96.1	38±92.8
31-45	66±134.8	36±63.1	76±105.7	22±45.9
46-60	21±31.1	27±61.3	111±121.7	34±61.5
Week 3				
0-15	575±208.0	580±173.6	645±145.3	566±144.7
16-30	260±186.0	290±198.7	267±148.7	73±109.7
31-45	98±88.9	68±88.7	102±87.4	85±151.6
46-60	144±193.6	37±97.9	107±112.8	106±132.5
Week 7				
0-15	525±215.3	443±130.8	503±68.5	450±145.9
16-30	164±144.6	188±183.9	227±120.5	85±100.2
31-45	171±194.4	93±118.5	106±112.2	103±111.5
46-60	110±103.2	81±108.0	160±149.5	69±74.0
Week 12				
0-15	636±227.3	569±182.9	547±87.9	502±172.4
16-30	249±145.4	190±123.7	161±147.0	101±141.2
31-45	113±112.7	125±114.0	143±150.4	76±89.1
46-60	163±210.6	58±97.0	104±106.2	82±60.0

a Data (n=10) were obtained from pages 648-679 of MRID 45546001.

Table 5e. Ambulatory activity subsession counts (mean±SD) in males [a]

Interval (min)	Dose (ppm)			
	0	5	20	80
Pretest				
0-15	132±60.1	96±40.4	140±65.8	115±63.1
16-30	15±20	14±14.9	14±21.0	14±12.8
31-45	13±41.4	13±26.3	13±27.5	1±3.5
46-60	0±1.0	14±43.5	3±7.8	15±45.5
Week 3				
0-15	197±69.2	171±80.0	187±73.0	166±73.2
16-30	53±43.9	53±33.6	62±42.9	37±40.7
31-45	22±51.9	36±40.1	33±40.6	8±9.5
46-60	20±41.0	8±14.3	6±13.6	1±1.3
Week 7				
0-15	128±42.1	141±72.9	134±70.8	88±40.3
16-30	37±31.6	38±20.7	47±23.9	7±8.4
31-45	38±28.4	14±22.0	32±33.0	7±14.8
46-60	11±19.4	12±22.6	22±43.2	28±48.2
Week 12				
0-15	146±41.3	133±48.1	143±42.1	112±63.1
16-30	29±17.4	30±28.6	27±24.4	22±20.5
31-45	21±31.5	15±15.8	11±15.0	6±8.6
46-60	8±14.0	5±7.5	5±8.6	6±16.0

a Data (n=10) were obtained from pages 648-679 of MRID 45546001.

Table 5f. Ambulatory activity subsession counts (mean±SD) in females [a]

Interval (min)	Dose (ppm)			
	0	5	20	80
Pretest				
0-15	139±71.6	132±37.9	129±87.6	130±66.2
16-30	39±62.8	1±1.3	12±26.8	11±33.5
31-45	24±58.8	8±20.5	22±36.3	6±14.8
46-60	2±4.4	8±19.4	31±35.6	10±19.7
Week 3				
0-15	199±83.3	221±88.0	252±57.1	216±60.1
16-30	76±69.5	100±81.0	81±62.3	23±47.0
31-45	26±25.5	19±26.1	31±29.6	29±60.3
46-60	42±58.5	12±38.3	31±44.1	39±53.9
Week 7				
0-15	164±80.1	155±45.4	187±35.0	151±61.6
16-30	44±47.5	53±58.0	61±47.5	21±30.0
31-45	43±62.4	26±40.4	40±48.9	32±39.8
46-60	27±34.5	20±29.7	49±50.0	15±20.4
Week 12				
0-15	234±106.9	200±79.5	195±34.6	177±66.5
16-30	78±59.9	49±44.8	46±52.9	29±56.9
31-45	33±37.6	34±40.4	47±54.1	21±24.4
46-60	50±92.5	15±31.2	25±23.6	21±17.2

a Data (n=10) were obtained from pages 648-679 of MRID 45546001.

E. SACRIFICE AND PATHOLOGY

1. Gross pathology - No treatment-related effect was observed on brain weight, length, or width (Table 6).

Table 6. Brain weights and measurements [a]

Parameter	Dose (ppm)			
	0	5	20	80
Males				
Weight (g)	1.94±0.062	1.96±0.095	1.99±0.111	1.97±0.094
Length (mm)	20.7±0.76	20.2±1.30	20.0±1.02	19.7±1.10
Width (mm)	14.4±0.57	14.9±1.48	14.5±0.78	13.9±1.18
Females				
Weight (g)	1.78±0.111	1.82±0.069	1.80±0.065	1.84±0.065
Length (mm)	20.0±1.23	19.9±1.23	19.8±0.75	19.1±1.17
Width (mm)	14.1±0.63	14.0±0.68	14.2±0.93	13.9±0.89

a Data (n=10) were obtained from pages 229-230 of MRID 45546001.

2. Neuropathology - No treatment-related neuropathological effects were observed. The lesions noted in the 80 ppm animals (*e.g.* minimal axonal degeneration of the sciatic and tibial nerves and subacute inflammation of the cervical spinal cord) were observed with equal frequency in the controls and/or are commonly observed in rats of this strain. No other lesions were observed in the neural or skeletal muscle tissues at any dose in either sex.

III. DISCUSSION and CONCLUSIONS

A. INVESTIGATORS' CONCLUSIONS - The investigators concluded that the LOAEL was 80 ppm, based on decreases in body weight, body weight gain, food consumption, and total and ambulatory motor activity in both sexes.

B. REVIEWER COMMENTS - No treatment-related effects were observed on mortality, clinical signs, brain weight, length, or width, or neuropathology. FOB testing revealed no treatment-related effects.

At 80 ppm, body weight was decreased throughout the study in males, and the decrease was significant ($p \leq 0.05$) at Weeks 3-6 (↓11-12%). Body weight gains at Weeks 0 to 1, 1 to 2, and 2 to 3 were decreased ($p \leq 0.05$) by 21-39%, and overall (Weeks 0-13) body weight gain was decreased (NS) by 12%. In females, body weight was consistently decreased (NS) by 3-6% from Week 2 until the end of the study. Body weight gain was generally decreased (NS) in females by 8-100% throughout the study, and was significant ($p \leq 0.01$) at Week 1 to 2 (↓53%). Food consumption was decreased ($p \leq 0.05$) at Weeks 0 to 1 and 2 to 4 by 12-13% in the males and at Weeks 0 to 6, 7 to 8, and 9 to 11 by 10-16% in females. Total motor activity was decreased throughout treatment in the males (↓36-40%; $p \leq 0.05$) and in the females (↓10-37%; NS). Additionally, ambulatory activity was decreased (NS) throughout treatment in the males (↓27-40%) and females (↓10-37%). Habituation was unaffected by treatment.

The LOAEL is 80 ppm (equivalent to 5.7/6.1 mg/kg/day in males/females), based on decreased body weight, body weight gains, food consumption, and total and ambulatory motor activity in both sexes. The NOAEL is 20 ppm (equivalent to 1.4/1.6 mg/kg/day in males/females).

The study is classified as **acceptable/guideline** and satisfies the guideline requirements (870.6200b) for an subchronic neurotoxicity study in the rat.

C. STUDY DEFICIENCIES - None

DATA FOR ENTRY INTO ISIS

Subchronic Neurotoxicity Study - rats (870.6200b)

PC code	MRID #	Study type	Species	Duration	Route	Dosing method	Dose range mg/kg/day	Doses tested mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day	Endpoint(s)	Comments
083601	45546001	subchr neurotox	rats	90 days	oral	diet	0.4-6.1	0/0, 0.4/0.4, 1.4/1.6, and 5.7/6.1 [M/F]	1.4	5.7	BW, BWG, FC, total and ambulatory motor activity	Toxicity

13544

R100923

Chemical:	**Fentin hydroxide**
PC Code:	**083601**
HED File Code	**13000 Tox Reviews**
Memo Date:	**06/18/2004**
File ID:	**TX0050309**
Accession Number:	**412-05-2000**

HED Records Reference Center
09/07/2004