microfiche

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

009776

OFFICE OF
PESTICIDES AND TOXIC
SUBSTANCES

MEMORANDUM

OCT 03 1992

SUBJECT: Carbaryl
Data Call-IN, 90-day Response

Submission No. S404776
Submission No. S420168
Chem. No. 056801
HED Project No. 2-0082
TOX Chem No.: 160

FROM: Ray Landolt 9/29/92
Review Section I
Toxicology Branch II
Health Effects Division (H7509C)

TO: Larry Schnaubelt, PM 72
Reregistration Branch
Special Review and Reregistration Division (H7508W)

THRU: Mike Ioannou, Section Head
Review Section I
Toxicology Branch II
Health Effects Division (H7509C) J. M. Ioannou 9/29/92
and
Marcia van Gemert, Branch Chief
Toxicology Branch II
Health Effects Division (TS-769C) M van Gemert 9/29/92

Registrant: Rhone-Poulenc Ag Company, letter of August 9, 1991

Action Requested: Response to Toxicology Data Requirement 158.340

1. Request to upgrade a One-Year Feeding Study in Beagle Dogs found deficient in Toxicology Review of October 29, 1987 (DER 006401), with the information provided in a 5-week dog feeding study.

2. Data waiver for Dog Teratology Study (83-3). This study was requested by Peer Review Committee for Reproductive and Developmental Toxicity, June 16, 1989.

i 34

Conclusion: 1. The 5 week dog feeding is acceptable and provides the data requested to upgrade the 1 year dog feeding study.

Considered together the 5 week and 1 year dog feeding studies satisfy the toxicology data requirement (83-1) for a nonrodent chronic feeding study. Based on the 5 week dog feeding study, the Toxicology Review of October 29, 1987 (DER 006401) of a One Year Oral Toxicity Study in Beagle Dogs is ammended as follows:

Cholinesterase

NOEL = 45 ppm (males 1.43 and females 1.54 mg/kg/day)

LEL = 125 ppm (males 3.37 and females 3.73 mg/kg/day) with a significant decrease in plasma (23%) and brain (20%) cholinesterase activity for females at this level.

With the supplementary information provided by the 5 week dog feeding study (MRID 420228-01), the 1 year dog feeding study (MRID 401667-01) may be upgraded from Supplementary to Minimum rather than Guideline because of the variability of brain cholinesterase values reported in the 1 year dog study.

2. The FIFRA 88 Review Committee of September 17, 1992 recommended that the Peer Review Committee for Reproductive and Develomental Toxicity consider the data waiver for a dog teratology study.

Consideration Given this Request:

A One Year Oral Toxicity Study in Beagle Dogs was reviewed October 29, 1987 with the conclusion that a cholinesterase NOEL was not demonstrated at the lowest level (125 ppm) fed. In this study carbaryl was fed at 0, 125, 400, and 1250 ppm to 6 animals/sex/dose for one year. Considerable variability in brain cholinesterase activity was reported in males and females fed all three dietary levels as compared to the control values. Additional information on historical control data for plasma, erythrocyte, and brain cholinesterase in the beagle dog was requested accompanied by the methodology for determination of cholinesterase activity (DER 006401, copy attached).

The requested cholinesterase historical control and methodology information was reviewed March 16, 1989 with the conclusion that a cholinesterase NOEL was not demonstrated. A significant ($p<0.05$) decrease in plasma (23%) and brain (20%) cholinesterase activity was reported in females fed the 125 ppm level for one year (DER 007086, copy attached).

Consideration Given this Request(con't)

Subsequently, the registrant met with members of Toxicology Branch II on July 12, 1989 to discuss the conduct of a 5 week dog feeding study composed of three dose levels (including 125 ppm level) plus controls to demonstrate a cholinesterase NOEL. In the one year dog study, plasma and erythrocyte cholinesterase activity peaked at five weeks and remained relatively constant for the duration of the study. This 5 week study was to simulate the one year feeding study in experimeral design except hematology, clinical chemistry, organ weights and histopathology were not required.

The 5 week study in dogs is without effects on plasma, erythrocyte, and brain cholinesterase activity in males and females at levels of 20, 45, 125 ppm as compared to the control values. By comparison females at the 125 ppm level in the 1 year study exhibited a significant ($p \leq 0.05$) decrease in plasma cholinesterase activity (19-23%) during weeks 5 to 26 as compared to the control values. Plasma cholinesterase activity was not significantly decreased for females by week 52 or for males during weeks 5 to 52 of the study at the 125 ppm level.

Brain cholinesterase activity in the 5 week study was comparable between the control and the three dietary levels tested. However, in the 1 year study brain cholinesterase activity was variable without an apparent dose response relationship in males and females fed the 125, 400, and 1250 ppm levels. The variability in the methodology for measuring brain cholinesterase was also apparent in the 2 week range finding study where dogs were dosed at 50, 100, 200, 400, 800, and 1600 ppm without an apparent dose response relationship in cholinesterase values (DER 006401).

The subchronic toxicity study in which dogs were fed carbaryl for 5 weeks does not satisfy the acceptance criteria for a guideline (82-1) nonrodent subchronic feeding study.

However, this 5 week feeding study is acceptable and does provide the information requested in toxicology review of October 29, 1987 (DER 006401) and March 16, 1989 (DER 007086) for the 1 year dog feeding study.

With the supplementary information provided by the 5-week dog feeding study (MRID 420228-01), the 1 year dog feeding study (MRID 401667-01) may be upgraded from Supplementary to Minimum, rather than Guideline, because of the variability of brain cholinesterase in the 1 year dog study.

Consideration Given this Request (con't)

Based on the 5-week dog feeding study, the Toxicology Review of October 29, 1987 (DER 006401) of a One Year Oral Toxicity Study in Beagle Dogs is ammended as follows:

Cholinesterase NOEL = 45 ppm (males 1.43 and females 1.54 mg/kg/day)

LEL = 125 ppm (males 3.37 and females 3.73 mg/kg/day) with a significant decrease in plasma (23%) and brain (20%) cholinesterase activity in females at this level.

009776

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D C 20460

OCT 29 1987

006401

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

Subject: One-Year Oral Toxicity Study in Beagle Dogs
Caswell No. 160

From: Ray Landolt
Review Section #1
Toxicology Branch / HED (TS-769)

To: Larry Schnaubelt, PM #12
Insecticide Rodenticide Branch
Registration Division (TS-767)

Thru: R. Bruce Jaeger, Section Head
Review Section #1
Toxicology Branch / HED (TS-769) 10/19/87

Recommendation: A cholinesterase no effect level cannot be demonstrated based on the information available. Additional data are needed.

Brain cholinesterase activity was variable in males and females fed all three dietary levels and depressed significantly ($p \leq 0.05$) for females as compared to the concurrent control values. Insufficient data is available to determine whether carbaryl has a compound related effect on plasma, erythrocyte or brain cholinesterase activity.
Additional data is requested from Hazleton Laboratories for the range of brain cholinesterase values observed in their dog colony. The methodology for the automated brain cholinesterase determinations used in this study are also requested.

016101

5

009776

006401

Reviewed By: Ray Landolt/Dr. Louis Kasza
Section I, Toxicology Branch (TS-769C)
Secondary Reviewer: R. Jaeger, Section Head
Section I, Toxicology Branch (TS-769C)

DATA EVALUATION REPORT

Study Type: One-Year Feeding - Dog

TOX Chem No.: 160

MRID No.: 401667-01

Date: March 18, 1987

Test Material: Carbaryl (99%) Technical

Synonyms: Sevin

Study No.: UA 400717

Sponsor: Union Carbide

Testing Facility: Hazleton Laboratories

Title of Report: One-Year Oral Toxicity Study in Beagle Dogs

Author: N. Nicki Hamada, Ph.D.

Conclusion:

Classification of Data: Supplementary

Deficiency:

A cholinesterase no effect level was not demonstrated in this study.

Systemic NOEL - 400 ppm
LEL - 1250 ppm with a significant ($p \leq 0.05$) decrease in body weight gain (50%) during weeks 0-5, a non significant decrease in food consumption (23%) and a significant ($p \leq 0.05$) decrease in albumin values during weeks 13, 26, and 52 by 9, 5, and 11%, respectively reported for this dietary level.

Cholinesterase NOEL and LEL cannot be determined without additional cholinesterase data requested on the historical control animals.

006401

Conclusions:

The variability of brain cholinesterase determinations in these dog studies is difficult to interpret without additional data. For example, brain AChE values are presented below for the one year study compared to the 2 week feeding study (copy attached). Control animals were not included in the two week feeding study

	Female Brain AChE		Male Brain AChE	
Dietary level ppm	1 year umol/g	2 week umol/g	1 year umol/g	2 week umol/g
0	9.3	-	11.3	-
50	-	10	-	5.6
100	-	7.0	-	6.1
125	7.2	-	9.7	-
200	-	7.1	-	6.4
400	7.0	6.7	7.7	6.7
800	-	4.3	-	4.3
1250	5.8	-	8.5	-
1600	-	4.2	-	4.6

The brain AChE activity for the females fed 50 ppm carbaryl in the two week study is higher than the control data in the one year study. The male data do not demonstrate a dose-related effect due to the variability in brain AChE levels. Data of this nature are extremely difficult to interpret without additional data on the analytical methodology for preparing and measuring brain cholinesterase, as well as variability in control data for beagles at Hazleton Laboratories. These data must be provided.

Plasma AChE activity for females fed 125 ppm carbaryl was significantly ($p \leq 0.05$) decreased and ranged from 19-23% depression compared to concurrent controls during the course of the study. When these same data were compared to each dog's baseline pre-dose data there was no apparent effect at 125 ppm. An individual animal serving as its own control, with relevent baseline data, is considered more meaningfull than a comparinson to a control group mean. However, as with brain AChE, Toxicology Branch requires additional information on historical control data for plasma and erythrocyte cholinesterase in beagle dog from Hazleton Laboratories. Such data should include the standard deviation for the sample size examined.

A. Materials:

1. Test Material - Carbaryl Technical is an off-white powder, identified as No. 12909 A-6 with a purity of 99 percent.

2. Test Animals - Twenty-four male and twenty-four female, 20 to 31-week-old purebred beagle dogs weighing 6.0 to 8.3 kg for males and 5.5 to 7.2 kg for females were divided into four dietary levels of 0, 125, 400, and 1250 ppm of six animals sex level.

B. Study Design:

1.

Test Group		Dose Level ppm	No. Males	No. Females
Control	1	0	6	6
Low	2	125	6	6
Mid	3	400	6	6
High	4	1250	6	6

2. Diet Preparation - A premix was prepared fresh weekly from which the required amount was used to prepare the dietary levels fed.

Weekly samples from each dietary level were pooled from weeks 1 to 4 and every 4th week thereafter and submitted for verification of concentration analysis. Carbaryl (99%) was stable in the diet for up to 3 weeks when either kept frozen or stored at room temperature.

Diet Concentration Analysis Over the 52-Week Period

Test Group		Target Dose Level ppm	Percent of Target
Control	1	0	Not detected
Low	2	125	97.12 - 108.9
Mid	3	400	98.0 - 108.2
High	4	1250	99.0 - 110.7

3. The basal diet (Purina Certified Canine Diet No. 5007) and tap water (via an automated watering system) were available ad libitum.

4. Statistical Analysis - Weekly body weights and food consumption, total (weeks 1-52) food consumption, clinical pathology (excluding cell morphology and urinalysis data), and organ weight data of the control group were compared statistically to the data from the same sex of the treated groups. Tests for homogeneity of variances and ANOVA were evaluated at the 5.0 percent two-tailed probability level. Control vs. compound-treated group mean comparisons were evaluated at the 5.0 percent two-tailed probability level. Cholinesterase activity was evaluated by single factor analysis of variance and Dunnett's t-test.

5. Quality Assurance - Patricia L. Range, March 18, 1987.

C. Methods and Results:

1. Observations - All dogs were observed twice daily for mortality and once daily for clinical signs of pharmacologic effects. All animals were housed individually.

   a) No deaths were reported during the duration of the study.

   b) The frequency of occurrence of pharmacotoxic signs for each dog over the 52-week period show a greater incidence among the high-dose females of emesis, lacrimation, salivation, and tremors.

2. Body weights were recorded at the beginning of each week. A significant ($p < 0.05$) decrease in body weight gain (50%) was reported for the high dose females during weeks 0-5 of the study. A slight decrease in body weight gain during weeks 6 to 9 of 4 percent and during weeks 28 to 36 of 6 percent was observed for the high-dose females. The terminal body weight for males fed the low, mid and high dietary levels were greater than the control body weights by 8, 9, and 15% respectively.

3. Food consumption was recorded individually at the end of each week. A (non-significant) decrease in food consumption was observed for females of the high-dose level of 21 and 22 percent during weeks 4 and 6 followed by an 18, 24, and 16 percent decrease during weeks 27, 28, and 29, respectively.

4. Ophthalmological examinations were performed on all dogs prior to the initiation of treatment and during the final week of the study using a slit lamp and an indirect ophthalmoscope. No dose-related ophthalmic abnormalities were observed.

5. Blood samples for hematology and clinical chemistry determinations were collected prior to the initiation of the study (week -2), then in the morning before feeding the dietary levels of carbaryl during weeks 13, 26, and 52 of the study.

a) Hematology - The following (X) parameters were determined:

X	Hematocrit (HCT)		Total plasma protein (TP)
X	Hemoglobin (HGB)	X	Leukocyte differential count
X	Leukocyte count (WBC)		Mean corpuscular HGB (MCH)
X	Erythrocyte count (RBC)		Mean corpuscular HGB conc. (MCHC)
X	Platelet count		Mean corpuscular volume (MCV)
X	Clotting time	X	Prothrombin time
X	Heinz bodies	X	Cell morphology

At the high dietary level, male leukocyte count was elevated above the control values during the pretreatment interval (16%), at the 13-week interval (13%), being significantly ($p < 0.05$) elevated during the 26th and 52nd week by 30 and 48 percent, respectively. The segmented neutrophil count of males fed the high dietary level was elevated above the control values at the pretreatment interval (22%), the 13-week interval (27%), the 26-week interval (44%), being significant ($p \leq 0.05$) at the termination of the study by 63 percent. A significant decrease in male clotting time was reported during the 26-week interval for the mid (17%) and high (21%) dietary levels.

b) Clinical Chemistry - The following (X) parameters were determined:

	Electrolytes:		Other:
X	Calcium	X	Albumin
X	Chloride		
	Magnesium	X	Blood urea nitrogen
X	Phosphorous	X	Cholesterol
X	Potassium	X	Globulins
X	Sodium	X	Glucose
	Enzymes	X	Total Bilirubin
X	Alkaline phosphatase	X	Total Protein
	Creatinine phosphokinase		Triglycerides
X	Lactic acid dehydrogenase	X	Albumin/globulin ratio
X	Serum alanine aminotransferase (also SGPT)	X	Creatinine (at 26 and 52 weeks only)
X	Serum aspartate aminotransferase (also SGOT)		
X	Methemoglobin		
X	Gamma glutamyltransferase		
X	Creatine kinase		
X	Direct bilirubin		

Female albumin values of the high dietary level were significantly ($p \leq 0.05$) decreased during weeks 13, 26, 52 by 9, 9, and 11 percent, respectively.

Blood creatinine levels were determined only for the 26 and 52 week intervals with the three dietary levels comparable to the control values for these two intervals.

c) Cholinesterase - Determined weekly over a 3-week period prior to treatment, then at 5, 13, 26, and 52 weeks for plasma and RBC activity. The three pre-treatment values for each subject were averaged to obtain a mean pre-treatment value for that subject. Blood samples taken for cholinesterase activity were collected two hours (midday) after the daily feeding period. Tissue for brain cholinesterase activity was taken, from the right hemisphere, two hours after the two hour feeding at the termination of the study. An analyzer was cited for the determination of cholinesterase activity.

1) The percent mean cholinesterase inhibition relative to the respective concurrent control values is presented in the following table.

Male - Mean Cholinesterase Activity (%) Inhibition

Group	Dietary Level ppm	Interval - Week 5 Plasma	5 RBC	13 Plasma	13 RBC	26 Plasma	26 RBC	52 Plasma	52 RBC	Brain
2	125	14	11	13	14	14	6	4	7	14
3	400	36*	23*	34*	28*	35*	19	30*	20	32
4	1250	66*	56*	57*	49*	59*	46*	58*	53*	25

Female - Mean Cholinesterase Activity (%) Inhibition

Group	Dietary Level ppm	Interval - Week 5 Plasma	5 RBC	13 Plasma	13 RBC	26 Plasma	26 RBC	52 Plasma	52 RBC	Brain
2	125	22*	13	23*	3	19*	9	12	7	20*
3	400	31*	34*	28*	29*	26*	29*	9	18	22*
4	1250	60*	38*	57*	29*	55*	37*	47*	30*	36*

*Significantly different from mean control value ($p \leq 0.05$).

2) The percent of mean cholinesterase inhibition relative to the pretreatment control values is presented in the following table.

Male - Mean Cholinesterase Activity (%) Inhibition

Group	Dietary Level ppm	Interval - Week 5 Plasma	5 RBC	13 Plasma	13 RBC	26 Plasma	26 RBC	52 Plasma	52 RBC	Brain
1	0	0	4	-1	5	-1	-3	5	-12	-
2	125	15	13	13	17	14	0	9	-5	-
3	400	36	28	32	33	33	17	32	13	-
4	1250	64	45	54	36	57	26	58	31	-

Female - Mean Cholinesterase Activity (%) Inhibition

Dietary Level ppm	Interval - Week 5 Plasma	5 RBC	13 Plasma	13 RBC	26 Plasma	26 RBC	52 Plasma	52 RBC	Brain
0	7	13	1	8	-2	-12	11	-9	-
125	15	4	11	13	3	0	8	2	-
400	37	22	30	31	26	17	21	9	-
1250	63	35	57	39	54	34	53	30	-

3) In Vitro cholinesterase studies- The I_{50} of carbaryl (99%) in the blood from untreated stock animals was determined to be around 10^{-6}M for erythrocyte and at around 10^{-5}M for plasma cholinesterase in both male and female beagle dogs. No apparent reason was given for the difference in sensitivity of the erythrocyte/plasma cholinesterase activity determined in vitro versus in vivo. Plasma cholinesterase activity is more sensitive than erythrocyte cholinesterase activity when measured in vivo.

6. Urinalysis - Urine samples were collected from cage pan runoff in the morning before the daily feeding period and on the day following the day blood was drawn for cholinesterase determination. The following (X) parameters were determined:

X	Appearance	X	Glucose
X	Volume	X	Ketones
X	Specific gravity	X	Bilirubin
X	pH	X	Blood
X	Sediment (microscopic)		Nitrate
X	Protein	X	Urobilinogen
		X	Reducing substances

No adverse findings were reported related to the dietary levels fed.

7. Sacarifice and Pathology After 52 weeks on study, all of the dogs were exsanguinated while under the influence of sodium thiamylal anesthesia. All animals were subjected to gross pathological examination, the (X) tissues were collected for histological examination, and the (XX) organs were weighed

	Digestive system		Cardiovasc./Hemat.		Neurologic
	Tongue	X	Aorta	XX	Brain
X	Salivary glands	XX	Heart		Periph. nerve
X	Esophagus	X	Bone marrow (sternum)	X	Spinal cord (3 level)
X	Stomach	X	Lymph nodes	XX	Pituitary
X	Duodenum	XX	Spleen	X	Eyes (optic n.)
X	Jejunum	X	Thymus		Glandular
X	Ileum		Urogenital	XX	Adrenals
X	Cecum	XX	Kidneys		Lacrimal gland
X	Colon	X	Urinary bladder	X	Mammary gland
X	Rectum	XX	Testes		Parathyroids
XX	Liver	XX	Epididymides	XX	Thyroids
X	Gallbladder	X	Prostate		Other
X	Pancreas		Seminal vesicle		Bone
	Respiratory	XX	Ovaries	X	Skeletal muscle
X	Trachea	X	Uterus	X	Skin
X	Lung				All gross lesions and masses
				X	Sciatic nerve

a) Gross Pathology

No gross pathological findings were observed related to the dietary levels fed.

b) Organ weights -

i. Absolute weights - A significant ($p < 0.05$) increase (24%) in absolute liver/gallbladder weight was reported for males fed the high dietary level. Male absolute pituitary weights were decreased (not significant) for the low, mid and high levels by 8, 15 and 22%, respectively as compared to the control values.

ii. Organ to body weight - A significant ($p < 0.05$) decrease (27%) in relative thyroid to body weight was reported for males of the high dietary level as compared to the control values. Liver to body weight ratio for males fed the high level was elevated (not significant) by 8% as compared to the controls. Male relative pituitary weights were decreased (not significant) for the low, mid and high dietary levels by 11, 22 and 33%, respectively as compared to the control values.

iii. Organ to brain weight - Liver to brain weight ratio for males fed the high level was elevated (not significant) by 21% as compared to controls. Male relative pituitary weights were decreased (not significant) for the low, mid and high dietary levels by 9, 18 and 27%, respectively as compared to the controls.

iv. The variations in male organ weights reported for liver, thyroid and pituitary are of questionable significance without collaborative effects for clinical chemistry and histopathological changes.

c. Microscopic pathology by Louis Kasza, Branch Pathologist, memorandum of June 15, 1987(copy attached).

"INTRODUCTION

Pathologic evaluation of the 'One-Year Oral Toxicity Study in Beagle Dogs with Carbaryl Technical' was forwarded to me by Dr. Farber and a particular request was addressed to me by the Section Head, Review Section #1, R. Bruce Jaeger, on June 3, 1987. The request was as follows:

'In connection with the memo from Dr. Farber requesting a review of the pathology data in the one-year dog study for carbaryl (received 5/18/87), please pay particular attention to the

urinary system. Earlier dog studies demonstrated an excretion of amino acids suggesting possible effects on the nephron. There was no evidence of similar effects in rats or mice. Your professional opinion is greatly appreciated.'

"MATERIALS AND METHODS

The summary incidence table was evaluated in Hazleton's report HLA Study No. 400-715. The incidences of pathologic changes were compared in test groups versus control group in male and female animals. The tabulated results are presented in Table 12 of the Company report. The grades of lesions were listed in each individual animal and the summarized results are tabulated. Grading the lesions, I followed the terminology of the investigating pathologist. In the attached table, I have summarized the results of the graded lesions.

"RESULTS

There are no significant differences in the pathologic changes between the control and test groups. It should be noted that all listed kidney lesions represent relatively minor changes which are frequently observed in dog kidneys. Furthermore, when the grades of lesions were compared in control and test groups, significant differences were also not found.

"CONCLUSION

Based on the presented data, compound-related changes in the urinary system in the presented one-year dog study could not be established."

009776
006401

16800:I:Landolt:C.Disk:KENCO:8/14/87:NeeCee:VO:SJH:dsg:aw:lisa:dsg:aw
R:16823:Landolt:C.Disk:KENCO:9/9/87:CB:VO:EK:CB

006401

16 17

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D C 20460

006401

009776

June 15, 1987

OFFICE OF PESTICIDES AND TOXIC SUBSTANCES

SUBJECT: Evaluation of Pathologic Findings in Urinary System. One-Year Oral Toxicity Study in Beagle Dogs with Carbaryl Technical. Caswell No. 160.

FROM: Louis Kasza
Branch Pathologist
Toxicology Branch/HED, TS-769

TO: R. Bruce Jaeger, Section Head
Review Section #1
Toxicology Branch/HED (TS-769)

INTRODUCTION

Pathologic evaluation of the "One-Year Oral Toxicity Study in Beagle Dogs with Carbaryl Technical" was forwarded to me by Dr. Farber and a particular request was addressed to me by the Section Head, Review Section #1, R. Bruce Jaeger, on June 3, 1987. The request was as follows:

> "In connection with the memo from Dr. Farber requesting a review of the pathology data in the one-year dog study for carbaryl (received 5/13/87), please pay particular attention to the urinary system. Earlier dog studies demonstrated an excretion of amino acids suggesting possible effects on the nephron. There was no evidence of similar effects in rats or mice. Your professional opinion is greatly appreciated."

MATERIALS AND METHODS

The summary incidence table was evaluated in Hazleton's report HLA Study No. 400-715. The incidences of pathologic changes were compared in test groups versus control group in male and female animals. The tabulated results are presented in Table 12 of the Company report. The grades of lesions were listed in each individual animal and the summarized results are tabulated. Grading the lesions, I followed the terminology of the investigating pathologist. In the attached table, I have summarized the results of the graded lesions.

009776

RESULTS

There are no significant differences in the pathologic changes between the control and test groups. It should be noted that all listed kidney lesions represent relatively minor changes which are frequently observed in dog kidneys. Furthermore, when the grades of lesions were compared in control and test groups, significant differences were also not found.

CONCLUSION

Based on the presented data, compound-related changes in the urinary system in the presented one-year dog study could not be established.

cc: T. Farber, TS-769
M. Landolt, TS-769
W. Burnam, TS-769

Attachment

006101

Data Call In

MRID #'s 401663-01 + 420227-01

Page 19 is not included in this copy.

Pages ___ through ___ are not included in this copy.

The material not included contains the following type of information:

_____ Identity of product inert ingredients.

_____ Identity of product inert impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

_____ The product confidential statement of formula.

_____ Information about a pending registration action.

✓ FIFRA registration data.

_____ The document is a duplicate of page(s) _____.

_____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.

GRADES OF KIDNEY LESIONS
CARBARYL. TECHNICAL STUDY IN BEAGLE DOGS

	MALE GROUPS					FEMALE
	1	2	3	4	5	6
DIAGNOSES						
Mineralization						
Minimal	5/6	6/6	6/6	5/6	5/6	5/6
Slight	0/6	0/6	0/6	0/6	1/6	1/6
Pigmentation						
Minimal	0/6	0/6	1/6	0/6	0/6	0/6
Slight	1/6	0/6	0/6	2/6	0/6	0/6
Regeneration						
Minimal	1/6	0/6	2/6	1/6	0/6	1/6
Slight	0/6	0/6	1/6	0/6	1/6	0/6
Mononuclear Cell Infiltration						
Minimal	0/6	0/6	1/6	0/6	2/6	2/6
Slight	0/6	0/6	1/6	0/6	0/6	0/6
Fibrosis						
Moderate	1/6	0/6	0/6	0/6	0/6	0/6
Fatty Changes						
Minimal	0/6	0/6	0/6	1/6	2/6	5/6
Slight	0/6	0/6	0/6	0/6	1/6	0/6
Moderate	0/6	0/6	0/6	0/6	0/6	0/6
Pelvic Inflammation						
Minimal	0/6	0/6	0/6	0/6	0/6	1/6

16900

009776

Two-Week Dose Range-Finding Oral Toxicity Study in Beagle Dogs. Hazleton Laboratories No. HLA 400-716, June 26, 1985, Acc. No. 401667-01.

Procedure

To select the dose levels for the 1-year dog feeding study, the technical material (99%) was administered in the diet to six groups of one male and one female (28- to 37-week-old) beagle dogs for 2 weeks at levels of 50, 100, 200, 400, 900, and 1600 ppm. No concurrent control group was designated for this study. "Criteria evaluated for compound effect included mortality, body weight gain, appearance, and behavior, food and compound consumption; clinical laboratory studies (hematology, clinical chemistry, and urinalysis); ophthalmologic examination, gross necropsy findings and organ weight data.

Results

1. Gross Observations - There was no mortality and no apparent changes in body weight gain among the test groups of either sex. Salivation was observed for the female fed the 1600 ppm level on day 2 of the study. In general, all female dogs fed dietary levels of 50, 100, 200, 400, 800, and 1600 ppm exhibited a decrease in food consumption by the conclusion of the study. However, no generalizations were apparent for the food consumption of males fed the corresponding dietary levels.

2. Hematology-A 42% decrease in male clotting time was observed for the 1600 ppm level at the termination of the study as compared to the pretest value.

3. Clinical Chemistry - No apparent dose related changes between the terminal and pretest values were observed.

4. Cholinesterase - Blood and brain tissue obtained for cholinesterase determinations were collected from unfasted dogs. The following table summarizes the plasma and RBC percent cholinesterase inhibition calculated from the mean pretreatment values* and the actual brain cholinesterase values for the six dietary levels.

*Mean pretreatment values were calculated from three samples collected at 4-day intervals prior to treatment.

Group	Dose Level ppm	Day 7 Plasma (%) Male	Day 7 Plasma (%) Female	Day 7 RBC (%) Male	Day 7 RBC (%) Female	Day 14 Plasma (%) Male	Day 14 Plasma (%) Female	Day 14 RBC (%) Male	Day 14 RBC (%) Female	Brain umol/g Male	Brain umol/g Female
1	50	11.5	14.6	11.4	15.1	20.6	13.5	25.7	9.3	5.6	10.0
2	125	16.5	19.6	11.6	9.8	16.5	19.6	10.1	9.8	6.1	7.0
3	200	21.1	29.9	8.9	27.6	25.3	23.7	16.1	28.1	6.4	7.1
4	400	35.4	44.1	32.8	40.4	36.6	41.2	35.9	42.6	5.7	6.7
5	800	47.3	54.6	41.5	50.0	45.9	49.5	52.3	48.9	4.3	4.3
6	1600	67.4	67.0	54.2	55.2	67.4	65.0	59.4	58.6	4.6	4.2

By inspection of the absolute values recorded for male brain cholinesterase activity in this study, it is apparent that the absolute values reported for the 50 through 1600 ppm dietary levels are within 4.3 to 6.4 umol/g range. No control animals were included in this study. When compared to the mean control values of 11.3 (8.8 - 17.3) umol/g reported for male brain cholinesterase activity at the termination of the 1-year study, the data presented for the 2-week study suggest an effect on brain cholinesterase inhibition at all levels tested in the males of this study. The mean female control value reported for brain cholinesterase activity at the termination of the 1-year study is 9.0 (7.3 - 10.9) umol/g, which suggests that the 50 ppm level may be the no-effect level for female brain cholinesterase inhibition in this study. Female brain cholinesterase mean percent values of the low level (125 ppm) in the 1-year dog feeding study were significantly ($p \leq 0.05$) different than the control values.

Plasma and RBC cholinesterase analyses were performed on day 13 from blood samples collected prior to feeding, at the end of the feeding period, and then at 2, 4, and 6 hours postfeeding from dogs of the 1600 ppm level. The peak for plasma and RBC cholinesterase inhibition was reported to be 4 hours after feeding the test material for both sexes. The percent plasma and RBC cholinesterase inhibition observed at the 4-hour interval was 58 and 56%, respectively, for males and 51 and 56%, respectively, for females.

5. Urinanalysis - No adverse findings were reported related to the dietary levels feed.

6. Ophthalmologic Examination - No compound or dose-related ophthalmic abnormalities noted.

7. Terminal Examination

a. No gross pathological observations related to the dietary levels fed were reported at necropsy.

b. No compound or dose-related changes in the absolute or relative organ weight change were observed.

009776

Reviewed By: Ray Landolt 4/28/92
Section I, Toxicology Branch II - H7509C
Secondary Reviewer: Mike Ioannou 4/28/92
Section I, Toxicology Branch II - H7509C

009776

DATA EVALUATION REPORT

Study Type: 5-Week Feeding - Dog (82-1)

Submission No. S404776
Project No. 2-0082
Chem No. 056801
Tox Chem No. 160
MRID No.: 420228-01

Test Material: Carbaryl (1-naphthyl N-methylcarbamate)

Date: March 28, 1991

Classification: Carbamate

Synonyms: Sevin

Study No.: HLA 656-152

Sponsor: Rhone-Poulenc AG Company

Testing Facility: Hazleton Laboratories

Title of Report: Subchronic Toxicity Study in Dogs with Carbaryl Technical

Author: N. Nicki Hamada, Ph.D.

Quality Assurance: Blain Wingard

Classification of Data: Supplementary to the One Year Dog Feeding Study

This subchronic toxicity study in which dogs were fed carbaryl for five weeks does not satisify the acceptance criteria for a guideline (82-1) subchronic feeding study in the dog.

However, this 5 week feeding study is acceptable and does provide information requested in Toxicology Review of October 29, 1987 (DER 006401) and March 16, 1989 (DER 007086) for a 1 year dog feeding study (MRID 401667-01) in which a a cholinesterase NOEL was not demonstrated at the lowest level fed (125 ppm).

Conclusion: Plasma, erythrocyte, and brain cholinesterase activity from feeding carbaryl to dogs at 20, 45, and 125 ppm was not significantly different from the respective control values over the 5 week period.

Cholinesterase NOEL = 125 ppm (males 5.83 and females 4.11 mg/kg/day)

A. Materials:

1. Test Material - Carbaryl Technical, a white powder of lot No. 87191, with a purity of 99.3% was used in this study.

2. Test Animals - Twenty-four male and twenty-four female, 6 month old purebred beagle dogs weighing 6.4 to 9.1 kg for males and 5.8 to 8.3 kg for females were divided into four dietary groups of 0, 20, 45, and 125 ppm of six animals/sex/level.

B. Study Design:

1. Allocation of Animals:

Test Group		Dose Level ppm	No. Males	No. Females
Control	1	0	6	6
Low	2	20	6	6
Mid	3	45	6	6
High	4	125	6	6

2. Selection of Dose Levels - To determine whether the marginal effects observed at the lowest level fed (125 ppm) in the 1 year dog study were reproducible or establish a NOEL for cholinesterase activity. Dose levels fed in the 1 year dog study are 125, 400, and 1250 ppm.

3. Animal Husbandry - All animals were housed individually with temperature (66 to 80°F) and humidity controlled to provide a uniform environment. A 12-hour light/dark cycle was provided. Water was available ad libitum. All dogs were vaccinated against distemper, hepatitis, leptospirosis, parainfluenza, parvovirus, and rabies prior to the three week acclimation period.

4. Diet Preparation - Fresh diets were prepared from a premix weekly and stored frozen for 1 week prior to feeding. All dogs were fed approximately 2.5 kg of feed per week between 8 and 10 AM each day, 7days/week for 5 weeks. Purina Certified Canine Diet No. 5007 was used as the control and for mixing the dietary levels fed.

5. Statistical Analysis - Body weight gain, total food consumption, and plasma, erythrocyte and brain cholinesterase data of the control group were compared statistically to the data from the same sex of the treated groups. Test for homogeneity of variances and ANOVA were evaluated at the 5.0 percent one-tailed probability level. Control versus compound-treated group mean comparisons were evaluated at the 5.0 percent two-tailed probability level.

C. Methods and Results:

1. Diet Analyses:

Test Group		Target Dose ppm	Percent of Target Dose		
			Homogeneity	Stability	Concentration
Control	1				
Low	2	20	92.4 to 99.9	89.9 to 94.9	90.8 to 99.8
Mid	3	45	---- -- ----	---- -- ----	92.9 to 105
High	4	125	93.7 to 102	---- -- ----	89.7 to 102

The dietary levels of carbaryl were homogenous, stable when either kept frozen or stored at room temperature and within acceptable concentration of the target dose.

2. Observations:

All dogs were observed twice daily for mortality and once daily for clinical signs of pharmacologic effects. In addition, detailed clinical observations were conducted once each week.

a. Clinical observations - Emesis and soft (mucoid) feces were observed among the control and test males and females prior to and/or following the presentation of food during weeks 2 to 5.

b. Mortality - No deaths were reported during the study.

c. Body weights were recorded initially then weekly through-out the study. There were no significant differences in body weight gain between the control and test groups fed carbaryl for 5 weeks.

d. Food consumption was recorded weekly throughout the study. There were no significant differences in food consumption between the control and test levels during the experimental period.

e. The mean dietary intake of carbaryl over the five week period is summarized in the following table from this report.

Group	Dietary level (ppm)	Mean Dietary Intake(mg/kg/day)	
		Male	Female
2	20	0.59	0.64
3	45	1.43	1.54
4	125	3.83	4.11

f. Ophthalmoscopic examinations were preformed on all dogs prior to the initiation of the study and during week 5 using indirect ophthalmoscope. Tropicamide ophthalmic solution (1% Mydriacyl®) was used as the mydriatic. No treatment related findings were observed between the test and control groups.

2. Clinical Findings:

Blood samples for plasma and erythrocyte cholinesterase determinations were collected prior to the initiation of the study on days -11, -8, and -5 then on days 14 and 32 of the study. At the termination of the study, approximately one gram sample was removed from the dorsal posterior right and left hemisphere of cerebrum of 2 animals/sex/group from the first 16 dogs necropsied on days 37, 38, and 39 for determination of brain cholinesterase activity.

Plasma, erythrocyte, and brain cholinesterase activity was determined by autoanalyzer. Samples were collected from unfasted animals approximately 2 hours from the termination of the feeding period.

A statistically significant ($p<0.05$) decrease in male plasma cholinesterase activity was reported on day 14 for the 20 and 125 ppm levels by 18 and 22%, respectively as compared to the concurrent control values. This significant difference in plasma cholinesterase activity was not apparent on day 32 of the study.

Plasma, erythrocyte, and brain cholinesterase activity from feeding carbaryl to dogs at 20, 45, and 125 ppm was not significantly different from the respective control values over the 5 week period. A summary table of the mean plasma, erythrocyte, and brain cholinesterase activity values reported in this study are attached.

Hematology, clinical chemistry and urinalysis parameters recommended in the testing guidelines of November 1989 for an acceptable study were not determined in this 5 week subchronic toxicity study.

3. Terminal Observations:

On completion of the experimental period (5 weeks) all animals were exsanguinated while under the influence of sodium thiamylal anesthesia. A complete gross necropsy was performed on each dog beginning approximately 2 hours from the termination of the feeding period.

No gross pathological findings were observed relative to the dietary levels fed. Organ weights were not recorded. Tissues were not subjected to histopathological examination.

Conclusions: This subchronic toxicity study in which dogs were fed carbaryl for 5 weeks does not satisify the acceptance criteria for a guideline (82-1) subchronic feeding study in the dog.

However, this 5 week feeding study is acceptable and does provide the information requested in toxiciology review of October 29, 1987 (DER 006401) and March 16, 1989 (DER 007086) for the 1 year dog feeding study (MRID 401667-01) in which a cholinesterase NOEL was not demonstrated at the lowest level fed (125 ppm).

Plasma, erythrocyte, and brain cholinesterase activity from feeding carbaryl to dogs at 20, 45, and 125 ppm were comparable to their respective control values over the 5 week period.

Cholinesterase NOEL = 125 ppm (males 3.83 and females 4.11 mg/kg/day)

Classification of Data: Supplementary to the One Year Dog Feeding Study.

TABLE 6
SUMMARY OF CLINICAL CHEMISTRY DATA
SUBCHRONIC TOXICITY STUDY IN DOGS WITH CARBARYL TECHNICAL

GROUP		PL-CHE - UNOL/ML DAY -11	-8	-5	14	32	RBC-CHE - UNOL/ML DAY -11	-8	-5	14
						MALES				
1	(0 PPM) MEAN	9.8	9.1	9.5	8.9	8.3	8.8	9.1	9.2	8.7
	S.D.	.53	.79	.63	.58	.80	1.05	1.23	1.29	1.2
	N	6	6	6	6	6	6	6	6	6
2	(20 PPM) MEAN	8.1	7.7	8.1	7.3*	7.1	7.8	8.1	8.2	7.8
	S.D.	1.09	1.19	1.25	1.27	1.08	2.38	2.24	1.51	1.8
	N	6	6	6	6	6	6	6	6	6
3	(45 PPM) MEAN	9.1	8.6	8.9	8.1	7.6	8.4	8.1	8.2	7.7
	S.D.	1.23	1.24	1.15	1.08	.82	1.34	1.39	1.48	1.4
	N	6	6	6	6	6	6	6	6	6
4	(125 PPM) MEAN	8.7	8.3	8.7	6.8*	7.0	8.8	8.2	8.4	7.8
	S.D.	2.08	2.18	2.14	1.15	1.73	1.80	1.60	1.55	1.0
	N	6	6	6	6	6	6	6	6	6
						FEMALES				
1	(0 PPM) MEAN	8.5	7.8	7.9	7.8	7.4	8.8	8.4	8.6	8.3
	S.D.	1.24	.64	.72	.88	.70	2.23	3.18	2.80	2.6
	N	6	6	6	6	6	6	6	6	6
2	(20 PPM) MEAN	8.8	9.1	8.9	8.8	8.0	8.4	8.1	8.8	7.8
	S.D.	1.00	1.50	1.54	1.35	1.12	3.02	1.99	2.77	2.8
	N	6	6	6	6	6	6	6	6	6
3	(45 PPM) MEAN	9.3	8.9	8.8	8.3	8.0	8.5	8.8	8.1	8.2
	S.D.	1.56	.78	1.04	1.24	1.13	1.38	1.40	1.69	1.5
	N	6	6	6	6	6	6	6	6	6
4	(125 PPM) MEAN	8.6	9.6	9.2	7.8	7.1	8.5	8.0	8.3	7.1
	S.D.	1.38	.98	.88	.55	.85	2.84	1.68	2.23	1.1
	N	6	6	6	6	6	6	6	6	6

* Significantly different from control value, $p \leq 0.05$.

REVIEWED

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

009776

MAR 16 1989

007086

009776 OFFICE OF PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: Carbaryl - One-Year Toxicity Study in Beagle Dogs

HED Project No. 9-0451

TOX Chem No.: 180

FROM: Fay Landolt
Review Section I
Toxicology Branch II - Herbicide, Fungicide and Antimicrobial Support
Health Effects Division (TS-769C)

TO: Dennis H. Edwards, Jr., PM 12
Insecticide-Rodenticide Branch
Registration Division (TS-767C)

THRU: Mike Ioannou, Acting Section Head
Review Section I
Toxicology Branch II - Herbicide, Fungicide and Antimicrobial Support
Health Effects Division (TS-769C)

and

Marcia van Gemert, Acting Chief
Toxicology Branch II - Herbicide, Fungicide and Antimicrobial Support
Health Effects Division (TS-769C)

Registrant: Rhone-Poulenc Ag Company
Letters of March 18, 1988 and November 15, 1988

Action Requested: Company response with cholinesterase historical control data and the brain cholinesterase assay method requested in the Toxicology Review of the One-Year Oral Toxicity Study in Beagle Dogs (F. Landolt, October 29, 1987).

Conclusions

1. The plasma, RBC and brain cholinesterase concurrent control values reported for the One-Year Oral Toxicity Study in Beagle Dogs (Study No. HLA 400-715) appear to coincide with the historical control values reported for the five, 1-year dietary dog studies conducted at Hazleton Laboratories between June 1984 and July 1987 (Tables attached). The five historical control studies were not identified by a study number. Since the study in question (HLA 400-715) was conducted during the period reported for these five studies; were the control values for HLA 400-715 included in the historical control values submitted?

2. The toxicology evaluation (October 29, 1987) of the One-Year Oral Toxicity Study (HLA 400-715) remains unchanged with the following amendment to the cholinesterase evaluation of this study.

   Classification of Data: Supplementary

   Deficiency: A cholinesterase no effect level was not demonstrated.

   Cholinesterase NOEL is less than 125 ppm.

   Cholinesterase LEL is 125 ppm with a significant ($p < 0.05$) decrease in plasma (23%) and brain (20%) cholinesterase values reported for female dogs fed this dietary level.

Recommendation:

The current ADI was calculated to be 0.1 mg/kg based on the 2-year rat feeding study no observable effect level of 10.0 mg/kg and an uncertainty factor of 100. The toxicity data base in support of tolerances for residues of carbaryl in or on food commodities is incomplete. A previously accepted nonrodent 1-year dog feeding study was evaluated and found deficient in the Carbaryl Registration Standard of March 30,1984. This 1-year dog feeding study is deficient for the lack of a cholinesterase no effect level at 125 ppm (3.1 mg/kg), the lowest level fed. The results of an acceptable dog feeding study would necessitate a re-evaluation of the ADI for carbaryl. Additional dietary levels below 125 ppm are required in a 1-year dog feeding study to demonstrate a cholinesterase NOEL for carbaryl.

JA

Historical Control Values (μMol/ml)

	Study 1	Study 2	Study 3	Study 4	Study 5
Males – Plasma Cholinesterase Wk. 52					
Mean	7.5	9.4	8.5	9.2	10.4
S.D.	1.20	0.87	0.93	1.36	1.00
N	6	6	6	6	5
Range	5.8-9.2	8.3-10.5	7.3-9.6	7.6-10.6	9.4-11.
Females – Plasma Cholinesterase Wk. 52					
Mean	8.2	8.4	9.0	10.0	10.2
S.D.	1.51	1.45	1.95	1.60	1.05
N	6	6	6	6	5
Range	6.1-10.0	6.7-9.9	6.5-12.3	8.0-11.9	7.3-12.
Males – Red Blood Cell Cholinesterase Wk. 52					
Mean	8.9	8.2	8.5	9.0	8.7
S.D.	3.10	1.14	1.85	2.98	0.92
N	6	6	6	6	5
Range	3.8-13.0	6.2-9.3	6.2-10.9	6.5-13.3	7.4-9.8
Females – Red Blood Cell Cholinesterase Wk. 52					
Mean	8.4	9.3	9.4	9.2	8.2
S.D.	1.83	1.42	1.55	2.48	1.84
N	6	6	6	6	5
Range	6.3-11.1	7.8-11.3	6.5-11.2	6.5-13.0	6.0-10.
Males – Brain Cholinesterase Wk. 53					
Mean	8.0	8.5	11.7	7.5	8.6
S.D.	0.83	0.73	1.97	1.21	0.66
N	6	6	6	6	5
Range	7.1-9.5	7.6-9.4	9.4-14.7	5.8-9.2	7.8-9.6
Females – Brain Cholinesterase Wk. 53					
Mean	7.7	9.0	11.9	8.2	7.9
S.D.	1.46	0.56	1.50	0.79	0.66
N	6	6	6	6	5
Range	6.4-10.2	8.2-9.6	8.9-13.2	7.3-9.1	7.1-8.1

TABLE 6
MEAN CHOLINESTERASE ACTIVITY VALUES
ONE YEAR ORAL TOXICITY STUDY IN BEAGLE DOGS WITH CARBARYL

GROUP AND DOSAGE LEVEL		PL-CHE UNITS/ML MEAN -3	-2	-1	5	13	26	52
								MALE
1	MEAN	8 4	8 6	8 5	8 5	8 6	8 6	8 1
0 PPM	S D	1 63	1 45	2 02	1 03	1 94	1 98	2 49
	N	6	6	6	6	6	6	6
2	MEAN	8 6	8 6	8 5	7 3	7 5	7 6	7 8
125 PPM	S D	1 19	1 36	1 02	1 04	1 16	1 05	1 31
	N	6	6	6	6	6	6	6
3	MEAN	8 4	8 6	8 1	5 4*	5 7*	5 6*	5 7*
400 PPM	S D	1 21	1 37	1 58	1 12	1 07	1 02	1 21
	N	6	6	6	6	6	6	6
4	MEAN	8 4	8 8	7 8	2 9*	3 7*	3 5*	3 4*
1250 PPM	S D	1 68	1 61	1 58	04	99	1 08	1 14
	N	6	6	6	6	6	6	6
								FEMALE
1	MEAN	8 7	8 4	8 8	8 1	8 6	8 9	7 7
0 PPM	S D	1 34	1 25	1 04	1 47	91	1 07	1 24
	N	6	6	6	6	6	6	6
2	MEAN	7 7	7 1	7 4	6 3*	6 6*	7 2*	6 8
125 PPM	S D	1 25	1 03	1 02	73	77	1 48	1 28
	N	6	6	6	6	6	6	6
3	MEAN	9 1	8 7	8 9	5 6*	6 2*	6 6*	7 0
400 PPM	S D	1 09	1 38	96	82	1 15	1 04	1 71
	N	6	6	6	6	6	6	6
4	MEAN	8 8	8 5	7 8	3 2*	3 7*	4 0*	4 1*
1250 PPM	S D	1 48	1 42	1 48	01	71	72	1 88
	N	6	6	6	6	6	6	6

* SIGNIFICANTLY DIFFERENT FROM MEAN CONTROL VALUE ($p \leq 0.05$).

TABLE 4 - CONTINUED
MEAN CHOLINESTERASE ACTIVITY VALUES
ONE-YEAR ORAL TOXICITY STUDY IN BEAGLE DOGS WITH CARBARYL

GROUP AND DOSAGE LEVEL		BOC-CHE UNOL/ML MEAN -3	2[a]	-1	5[b]	13[b]	26	52
							MALE	
1	MEAN	7 4	7 9	7 6	7 3	7 2	8 0	8 5
0 PPM	S D	1 70	1 36	1 46	1 62	1.43	1 21	1 77
	N	6	6	6	6	6	6	6
2	MEAN	7 2	7 8	7 4	6 5	6 2	7 5	7 9
125 PPM	S D	1 53	1 48	1 45	1 23	1.46	1 18	1 58
	N	6	6	6	6	6	6	6
3	MEAN	7 7	8 2	7 5	5 6*	5.2*	6 5	6 8
400 PPM	S D	1 18	1 69	1 59	98	61	94	89
	N	6	6	6	6	6	6	6
4	MEAN	5 7	5.9	5 6	3 2*	3 7*	4 3*	4.8*
1250 PPM	S D	1 12	1 25	81	73	84	87	55
	N	6	6	6	6	6	6	6
							FEMALE	
1	MEAN	8 8	9 6	9 4	10 5	8 6	10 4	10 0
0 PPM	S D	1 61	2 87	2 17	1 99	1.85	1 66	2 83
	N	6	6	6	6	6	6	6
2	MEAN	8 9	10 1	9 3	9 1	8 3	9 5	9 3
125 PPM	S D	1 09	1 74	1 23	1 55	1 64	1 24	1 26
	N	6	6	6	6	6	6	6
3	MEAN	8 7	9 2	8 9	6 9*	6 1*	7 4*	8 2
400 PPM	S D	1 22	89	1 03	76	68	75	1 89
	N	6	6	6	6	6	6	6
4	MEAN	9 4	10 5	10 8	6 5*	6 1*	6 6*	7 8*
1250 PPM	S D	77	30	94	79	76	99	64
	N	6	6	6	6	6	6	6

[a] RANK-TRANSFORMED FEMALE DATA ANALYZED.
[b] LOG[10] TRANSFORMED FEMALE DATA ANALYZED.
* SIGNIFICANTLY DIFFERENT FROM MEAN CONTROL VALUE ($p \leq 0.05$).