2-25-92

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

009328

OFFICE OF
PESTICIDES AND TOXIC
SUBSTANCES

MEMORANDUM

SUBJECT: Prometon Review of a 1-Year Dog Feeding Study
EPA ID No. 080804-000100 Record No. S394514
MRID No. 400979-01
HED Project No. 1-1026
Tox Chem No. 096

FROM: Vivian A. Williams, M.S.
Toxicology Section II
Toxicology Branch I
Health Effects Division (H7590C)

TO: Jay Ellenberger, PM 50
Reregistration Division (H7508W)

THRU: Joycelyn Stewart, Ph.D.
Section Chief
Toxicology Branch I
Health Effects Division (H7590C)

Karl Baetcke, Ph.D.
Branch Chief
Toxicology Branch I
Health Effects Division (H7590C)

Conclusion:

This study showed that following oral administration of **Prometon** (gelatin capsules) to normal, healthy **beagle dogs** of both sexes for **one year** at dose levels of 0, 5, 20 and 50 mg/kg/day, emesis, diarrhea, lethargy, mydriasis, and ptosis occurred at the two high dose groups. The emesis was of particular concern because it was so prevalent in the high dose animals during study days 1-5 that adjustments to the original dosing regimen were made. The original dose levels of 0, 15, 50 and 90 mg/kg/day were lowered to 0, 5, 20, and 50 mg/kg/day in an attempt to overcome this finding; dosing was then suspended for 2 days and reinstated on day 8 for the full 52 weeks. Although the testing facility felt that these adjusted dose levels were sufficient to allow for continuance of the test, there is concern that the emesis, which still persisted in all dose groups (especially in the two high dosed groups) until study

termination, may have lead to an improper characterization of prometon's toxicity. Furthermore, in all dose groups inclusive of controls, diarrhea was noted and it, too, occurred in an increased frequency in the 50 and 20 mg/kg/d male and female dogs.

In addition to the emesis and diarrhea, there was an effect on body weight measurement and/or food consumption and clinical condition, wherein food consumption of high-dose males and females was depressed throughout the dosing period. Mean body weights and body weight gains for the 52-week study period were depressed in the high dose males and mid- and high-dose females. Body weights of females were depressed to a greater extent when compared to those of the males. Reductions occurred in total protein, globulin and cholesterol levels of high-dose males at weeks 13, 26, and 52 (the scheduled times for clinical lab tests). The reduced body weights, reduced body weight gains and the clinical biochemistry profile may be related to the emetic episodes, or, these symptoms may be representative of prometon's activity, if any of the compound did indeed get in and was retained in an appreciable amount. Two other triazine compounds, simazine and atrazine, when tested (in rats and dogs) have demonstrated body weight reductions and reductions in total globulins but no emesis. Due to the question concerning administration of the chemical as well as retention of the chemical in this dog study, a comparison of toxicities from this study to other triazine studies may not be relevant.

It should be noted that those control and high dose dogs (both sexes) which were held for an additional 4 weeks as recovery animals showed body weight recovery during weeks 53 to 57.

Since there is a lack of a more marked toxic response, it is not conclusive that the effects seen resulted from administration of prometon. Although it appears that prometon did not result in any compound-related changes in urinalyses or organ weights and there were no compound-related pathological (or histopathological) observations or no unscheduled deaths and unscheduled sacrifices in this study, the uncertainty of dose retained due to the emetic episodes seen at all dose levels limits the true characterization of the chemical. Additionally, **no NOEL has been established** for this study since emesis and diarrhea were observed in all groups tested. Based on these factors, this chronic dog study is classified as **Supplementary**.

Although it is stated that the animals were normal and healthy at study initiation, Toxicology Branch I has concerns for the persistent diarrhea and vomiting and, consequently, for the results of this study because these incidences are not normally seen in healthy animals. Hence, it is requested that the company provide information which will explain the repeated occurrences of these two effects at all dose levels (inclusive of the controls) and any other information which may be relevant in

assessing the potential toxicity of prometon in the dog. This may include but not be limited to the range-finding data that were utilized in setting the dose levels for this study and the rationale for using capsule administration of the chemical instead of the more common method of direct administration of the chemical in the food.

In the reviewer's attempt to better characterize the noted findings, attention was specifically directed towards examining the individual animal data as presented in the sponsor's submitted package, but the limitation to this approach was that the report of the clinical signs (for the emesis and diarrhea) was provided only by its weekly incidence. A more in depth account by the reviewer was not possible since the daily occurrence of the vomiting (or its incidence of daily episodes) in addition to its relationship to the time of dosing was not provided. As a result, the dose received and retained becomes questionable.

Overall, there is a request for information used in determining the dose levels for this study, an explanation for the frequent emesis and diarrhea at all dose levels and evidence that proves receipt and retention of the intended doses.

If the sponsor can satisfactorily address these concerns by providing the appropriate information, this study could be considered for an upgrade in classification.

Attached is the Data Evaluation Report for the 1-year oral dog study which tested the chemical Prometon. Please note that **there is an addendum to the contractor-generated DER.** Although this study, when reviewed by the contractor was classified as Core Minimum, an additional review lead to an increase in concern for the noted findings and it was determined that no NOEL has been established. This study is currently classified as **Core Supplementary.**

009328

Reviewed by: Vivian Williams
Section II, Tox Branch I (H75090)
Secondary Reviewer: Joycelyn Stewart, Ph.D. 7/14/92
Tox. Branch I
Date: January 29,1992

ADDENDUM

Although the attached DER states that this chronic dog study is Core Minimum, an additional review of the data has significantly increased the concern for the emesis, thereby downgrading the classification to **Supplementary.** Since the emetic effect was noted in all the experimental groups including the controls, **no NOEL is established.**

Even though the study states that the animals were healthy when this chronic test started, there is concern for the persistent emesis and diarrhea. To demonstrate the frequency of the vomiting, its weekly incidence in the control animals is as follows:

Control Male Dogs

Animal #	Week of Occurrence *
113	Not seen
391	Not seen
422	18
462	33
649	14
695	Not seen
711	24
795	11 & 13

Control Female Dogs

	Week of Occurrence *
131	8, 13 & 22
143	16, 43 & 49
351	Not seen
546	Not seen
556	7, 12, 14, 22, 32, 48, & 52
915	26, 40 & 52
922	52
977	12, 42 & 52

*Animals were examined on a daily basis, however the incidence of emesis is listed in the data according to its weekly occurrence.

0004

Although the emesis was seen in the control animals, it was most prevalent in the high dosed animals, having a weekly incidence which is shown below:

High Dose Male Dogs

Animal #	Week of Occurrence
123	1-11, 14, 15, 17, 19-25, 31-36, 39-42, 45, 46, 48, 49 & 52
233	1, 3, 22, 32, 34, 36, 39, 40, 41, 45, 46, 49 & 51
300	1-3, 5, 7, 8, 10, 11, 13-16, 19-21, 23, 27, 28, 30, 33-37, 39-43, 45-51 & 53
397	1-5, 10, 11, 14, 16, 22-24, 26, 27, 31, 34, 37, 41, 42, 51 & 53
563	1-3, 5, 6, 8-15, 17-26, 28 & 53
733	1, 3, 5, 7, 14, 15, 20, 25, 27, 28, 33-35, 37, 39, 41,44, 49 & 53
734	1-7, 9, 11-14, 16-18, 21,23, 25, 27-29, 31-34, 36-49 & 52
991	1-3, 6-8, 13, 20-22, 28-30, 32, 35-39, 46, 47 & 51

High Dose Female Dogs

031	1, 2, 4-6, 13, 18-20, 23-25, 29, 33-36, 39, 40, 42, 45-48, 50-52 & 57
032	1-6, 9, 12, 15, 17-19, 23-25, 29, 31-34, 37-39, 45 & 47-52
334	1-9, 12-15, 17-25, 27, 28, 30-35, 39-50, 52, 53 & 57
370	1, 3, 4, 14, 25, 36, 39, 41, 45 & 52
398	1, 3-5, 7-10, 12, 15, 19, 20, 22, 27, 33-35, 40,42, 44, 46-49 & 51-53
772	1, 6, 10, 14, 15, 19, 36, 37, 39, 44, 48, 50 & 54
865	1-3, 5, 22, 30, 35, 45-47, 50 & 53
870	1, 6, 12, 15, 16, 20, 23, 25-27, 30-32, 43, 45 & 52

As previously stated, there is a lack of a more marked toxic response in this study in that prometon did not result in any compound-related changes in urinalyses or organ weights and there were no compound-related pathological (or histopathological)

observations or no unscheduled deaths and sacrifices. The question of whether the effects seen were due to administration of the chemical as well as whether the animals retained the intended doses due to the emetic episodes (seen at all dose levels inclusive of the controls) limits the true characterization of the chemical.

Also, from the review of the available individual animal data for clinical signs, diarrhea was observed to be a frequent occurrence for which there is concern. It, too, occurred at all dose levels (inclusive of the controls) and its frequency appears to be greater in the two highest dose groups (both sexes) than in the other groups. The frequency of diarrhea in both sexes of the control and low dose groups appears to be about the same. Considering the magnitude/frequency of the diarrhea and vomiting throughout this study, the sponsor has provided no explanation as to why either of these findings occurred. **The recurrence of the diarrhea and emesis at all dose levels prevents the establishment of a NOEL for this study.**

This study is classified as **Core Supplementary**. If the sponsor can provide information that will sufficiently clarify the concerns which have been listed regarding compound administration and dose retention and upon the determination of a NOEL, the classification of this study could be considered for an upgrade.

CONFIDENTIAL BUSINESS INFORMATION
DOES NOT CONTAIN
NATIONAL SECURITY INFORMATION (EO 12065)

009328

EPA No.: 68D80056
DYNAMAC No.: 367-E
TASK No.: 3-67E
October 3, 1991

DATA EVALUATION RECORD

PROMETON

Chronic Oral Toxicity Study in Dogs

APPROVED BY:

Robert J. Weir, Ph.D.
Program Manager
Dynamac Corporation

Signature: [signature]

Date: 10-3-91

0007

009328

EPA No.: 68D80056
DYNAMAC No.: 367-E
TASK No.: 3-67E
October 3, 1991

DATA EVALUATION RECORD

PROMETON

Chronic Oral Toxicity Study in Dogs

REVIEWED BY:

Margaret E. Brower, Ph.D.
Principal Reviewer
Dynamac Corporation
Signature: Margaret Brower
Date: October 3, 1991

William L. McLellan, Ph.D.
Independent Reviewer
Dynamac Corporation
Signature: William L. McLellan
Date: October 3, 1991

APPROVED BY:

Nicolas P. Hajjar, Ph.D.
Department Manager
Dynamac Corporation
Signature: Nicolas P. Hajjar
Date: October 3, 1991

Vivian Williams, Ph.D.
EPA Reviewer
Review Section IV
Toxicology Branch I
(H-7509C)
Signature: V. Williams, MS
Date: 10/8/91

Joycelyn Stewart, Ph.D.
EPA Section Head
Review Section IV
Toxicology Branch I
(H-7509C)
Signature: Joycelyn Stewart
Date: 7/11/92

DATA EVALUATION RECORD

GUIDELINE § 83-1

STUDY TYPE: Chronic oral toxicity study in dogs.

MRID NUMBER: 400979-01.

TEST MATERIAL: Prometon; 2,4-bis (isopropylamino)-6-methoxy-s-triazine.

SYNONYM: Pramital.

STUDY NUMBER: 100-84.

SPONSOR: Agricultural Division, Ciba-Geigy Corporation, Greensboro, NC.

TESTING FACILITY: Pharmaceuticals Division, Ciba-Geigy Corporation, Summit, NJ.

TITLE OF REPORT: Prometon - One Year Oral Administration to Dogs.

AUTHORS: Breckenridge, C., and Green, J.

REPORT ISSUED: December 15, 1986.

CONCLUSIONS:

Prometon was administered in gelatin capsules to groups of five male and five female beagle dogs for 1 year at dose levels of 0, 5, 20, or 50 mg/kg/day. Three additional control and high-dose animals/sex were dosed for 52 weeks and held for 4 additional weeks as recovery animals. At study initiation, the highest dose tested was 90 mg/kg/day for the first 5 days. Because severe emesis occurred at the high dose, dosing was suspended for 2 days for all animals and reinitiated at day 8 at the lower dose levels. No deaths occurred. The incidence of emesis, diarrhea, lethargy, mydriasis, and ptosis was increased in high-dose males and females. Salivation was increased in all dosed males and high-dose females. Tremors were exhibited in high-dose males and in one female/group at all dose levels.

Prometon had a significant toxicological effect on body weight measurement and/or food consumption and clinical condition at doses of 20 and 50 mg/kg/day. Mean body weights and body weight gains for the 52-week study period were depressed in high-dose males and mid- and high-dose females. Weights of females were depressed to a greater extent when compared to those of males. Body weights of high-dose males and females recovered during weeks 53 to 57. The food consumption of high-dose males and females was depressed throughout the dosing period. Reductions occurred in total protein, globulin, and cholesterol levels of high-dose males at weeks 13, 26, and 52. There were no compound-related changes in urinalyses, organ weights, or pathological observations. Based on clinical observations and the depression of body weight, the LOEL is 20 mg/kg/day, and the NOEL is 5 mg/kg/day. The maximum tolerated dose is 50 mg/kg/day.

Classification: Core Minimum. This study meets the minimum requirements of Guideline 83-1, Chronic Toxicity in Beagle Dogs.

A. MATERIALS:

1. Test Compound: Prometon technical; description: white powder; batch No.: FL 821847; purity: 97%.

2. Test Animals: Species: dog; strain: beagle; age: 24 to 28 weeks at study initiation; weight: males--7.6 to 9.8 kg, females--6.5 to 9.3 kg; source: Marshall Research Animals, North Rose, NY.

B. STUDY DESIGN:

1. Animal Assignment: Following 6 weeks of acclimation, animals were assigned to the following test groups via computer randomization:

Test Group	Dose in Diet[a] (mg/kg/day)	Main Study (53 weeks) Males	Main Study (53 weeks) Females	Recovery Sacrifice[b] (57 weeks) Males	Recovery Sacrifice[b] (57 weeks) Females
1 Control	0	5	5	3	3
2 Low (LDT)	5	5	5	0	0
3 Mid (MDT)	20	5	5	0	0
4 High(HDT)	50	5	5	3	3

[a]The test material was initially administered at doses of 0, 15, 50, or 90 mg/kg/day for 5 days. Because emesis occurred at 90 mg/kg/day, dosing was suspended for 2 days in all animals and reinitiated on day 8 at doses of 0, 5, 20, or 50 mg/kg/day for 52 weeks.

[b]Three control and high-dose dogs/sex were held for 4 additional weeks as recovery animals.

Dogs were treated for intestinal parasites and were vaccinated against distemper, hepatitis, leptospirosis, Bordetella-parainfluenza, and oral papilloma by the supplier. Dogs were housed in an environmentally controlled room with a temperature of 20 ± 2°C, humidity of 50 ± 20%, and a 12-hour light/dark cycle.

2. Dosing Preparation and Administration: The test material was prepared in gelatin capsules at doses of 15, 50, and 90 mg/kg/day during study days 1 to 5. From study days 8 to study termination, capsules were prepared to contain doses of 5, 20, and 50 mg/kg. Dosing was based on the projected body weight at midweek. A maximum of 2 weeks elapsed between capsule preparation and administration. Three empty gelatin capsules were administered daily to each control dog.

Capsules were administered during late morning hours with approximately 1 hour separating the dosing of each capsule when multiple capsules were administered (control, mid-, and high-dose groups). Beginning on study day 8, a portion of the 400-g food allotment was fed to each animal 30 minutes before capsule administration. The doses of 0, 5, 20, or 50 mg/kg/day were administered for 52 consecutive weeks. Stability analyses of the test material were not conducted.

Results: Protocol deviations indicate that the five mid-dose females received 10 mg/kg rather than 20 mg/kg on one study day, and one high-dose male (animal No. 397) received 83.9 mg/kg rather than 90 mg/kg for 5 study days. In addition, one low-dose male received 4.9 mg/kg rather than 5 mg/kg for 3 study days. These dose changes were indicated to have no adverse effect on the study.

3. Food and Water Consumption: Animals were offered 400 g of dry, pelleted, standard dog diet daily for 3.5 hours and water ad libitum.

4. Statistics: The following procedures were utilized in analyzing the numerical data. Body weights, food consumption, clinical laboratory parameters, and organ weight data were examined for equality of group means at predosing using the F-test. The T-test was used to compare data from treated and untreated groups during the study. The trend test was used to detect differences among treatment groups with respect to average response and rate of change of response. Fisher's Exact test was used to compare histological observations.

5. Quality Assurance: A quality assurance statement was signed and dated December 15, 1986.

C. METHODS AND RESULTS:

1. Observations: Animals were inspected three times daily on weekdays and twice daily on weekends and holidays for signs of morbidity and mortality. Physical examinations were performed prior to dosing and monthly thereafter. Tissue mass palpations, heart rates, and rectal temperatures were also measured at these times.

Results: No deaths were reported during the study period. Clinical findings of dosed animals are summarized in Table 1. The study authors reported that there was a dose-related increase in the incidence of emesis during study days 1 to 5 when dogs were administered 15, 50, or 90 mg/kg/day. Emesis was increased to a greater extent in males; the total incidence was 0/40, 5/25, 19/25, and 32/40 in control, low-, mid-, and high-dose males, respectively, over a 5-day period (8 animals/group) and 0/40, 2/25, 12/25, or 20/40 in control, low-, mid-, and high-dose females, respectively, over the same time. The incidence of emesis slightly decreased following the dose reduction to 5, 20, or 50 mg/kg/day on study day 3. In addition, mid- and high-dose animals were administered the test material in multiple subdoses separated by feeding periods

TABLE 1. Clinical Findings in Dogs Administered Prometon for 53 Weeks[a,b]

	Dose Level (mg/kg/day)							
	Males				Females			
Clinical Finding	0	5	20	50	0	5	20	50
Ataxia	0	0	0	1	0	0	0	0
Cachexia	0	0	0	1	0	0	0	0
Dermatitis	0	1	1	2	1	0	1	4
Diarrhea	5	5	5	8	7	5	5	8
Emesis	5	5	5	8	6	4	5	8
Fecal changes (e.g., blood, mucous, soft, colored)	8	5	5	8	8	5	5	8
Hyperactivity	0	0	0	0	0	0	1	1
Hypoactivity	1	0	1	1	0	1	0	1
Hypothermia	0	0	0	1	0	0	0	0
Lethargy	1	0	1	8	1	0	1	7
Mydriasis	0	0	0	8	0	0	3	8
Ptosis	0	0	0	4	0	0	1	7
Salivation	0	2	2	3	1	0	1	5
Tissue mass	0	0	0	0	0	0	0	1
Tremors	0	0	0	3	0	1	1	1
Vulva-bloody discharge					1	0	1	1

[a]Based on eight animals/dose/sex in control and high-dose groups and five animals/dose/sex in low- and mid-dose groups.

[b]The incidence of clinical signs for week 1 (doses 15, 50, and 90 mg/kg/day) are included.

in an attempt to reduce the emesis. However, emesis continued throughout the study, especially in high-dose males and females and to a lesser extent in mid-dose animals. Emesis was exhibited at the greatest frequency in high-dose males (13 to 44 weeks in 8/8 animals) when compared to concurrent controls (1 to 2 weeks in 5/8 animals).

The incidence of diarrhea was increased at the high dose when compared to concurrent controls. Control and dosed animals also exhibited changes in fecal composition and color. High-dose males and females exhibited a high incidence of lethargy, mydriasis, and ptosis. The incidence of salivation was increased at all dose levels in males and at the high dose in females. Tremors were exhibited in 3/8 high-dose males, 1/5 low- and mid-dose females, and 1/8 high-dose females. High-dose male No. 734 exhibited decreased body weight and food consumption at study week 43; during weeks 44 to 47, the dog became lethargic, hypothermic, and cachectic with salivation, emesis, fecal changes, tremors, hypoactivity, tachypnea, lacrimation, ataxia, ptosis, miosis, dehydration, and conjunctivitis. The dog began to improve at study week 48. Two males with dermatitis were replaced prior to dosing, and one female with dermatitis was replaced on day 19. A few dosed and control animals were isolated and treated for dermatitis during the study (one high-dose male, and one mid- and two high-dose females); these dermal changes were not considered to be related to dosing. Other findings were also not considered to be related to dosing. The study authors reported that slight changes in body temperature and heart rates in dosed animals were not considered to be compound-related.

2. Body Weight: Dogs were weighed prior to dosing and weekly thereafter.

Results: Representative data on mean body weights and body weight gains are summarized in Tables 2 and 3, respectively. Body weights of high-dose males were significantly depressed from study weeks 4 to 6, 11 to 28, 33, and 37 to 43 ($p < 0.05$), and during study weeks 7 to 10, and 44 to 53 ($p < 0.01$). The body weight loss of one high-dose male (animal No. 734) was excessive from study week 45 to 50. Percent weight gains of high-dose males were significantly ($p < 0.01$) depressed (from 29 to 63%) throughout the study when compared to concurrent controls; weight gains of mid-dose males were slightly depressed at weeks 0-2, 0-4, 0-5, 0-7, and 0-8 when compared to concurrent controls. Body weights and body weight gains of low-dose males were consistently higher than concurrent controls; the weights of these animals were reported to be "comparable" to control weights by the study authors. Body

TABLE 2. Mean Body Weights at Selected Intervals in Dogs Administered Prometon for 53 Weeks[a]

Dose Level (mg/kg/day)	Mean Body Weight (kg ± SE) at Study Weeks: 0	13	26	53	56[b]
			Males		
0	8.6 ± 0.20	11.0 ± 0.34	11.8 ± 0.32	12.5 ± 0.35	12.3 ± 0.20
5	9.0 ± 0.25	11.5 ± 0.32	12.6 ± 0.35	13.5 ± 0.57	--
20	8.5 ± 0.41	10.6 ± 0.39	11.8 ± 0.41	12.1 ± 0.43	--
50	9.0 ± 0.14	10.0 ± 0.20*	10.8 ± 0.29*	10.8 ± 0.30**	11.6 ± 0.68
			Females		
0	7.7 ± 0.22	10.0 ± 0.36	11.3 ± 0.59	12.4 ± 0.78	12.4 ± 0.55
5	8.1 ± 0.25	9.6 ± 0.44	10.6 ± 0.42	11.1 ± 0.55	--
20	7.9 ± 0.55	9.0 ± 0.50	9.6 ± 0.51*	9.6 ± 0.56**	--
50	7.9 ± 0.29	8.4 ± 0.37**	9.0 ± 0.36**	9.2 ± 0.31**	9.5 ± 0.70*

[a]Based on eight controls and eight high-dose animals/sex and five low- and five mid-dose animals/sex.

[b]Based on three control and three high-dose recovery animals/sex held for 4 additional weeks.

*Significantly different from control at $p < 0.05$.

**Significantly different from control at $p < 0.01$.

TABLE 3. Mean Body Weight Percent Gain (± SE) at Selected Intervals in Dogs Administered Prometon for 53 Weeks[a,b]

Dose Level (mg/kg/day)	Mean Body Weight Percent Gain Compared to Week 0 at Study Weeks: 1	13	26	53	57[c]
			Males		
0	1.80 ± 0.98	28.20 ± 2.80	38.09 ± 3.05	45.65 ± 4.04	39.41 ± 6.66
5	0.65 ± 0.96	28.30 ± 2.27	39.92 ± 5.10	50.51 ± 9.13	--
20	-0.23 ± 0.86	25.32 ± 3.00	39.13 ± 4.23	43.61 ± 4.22	--
50	-1.28 ± 0.69*	10.55 ± 1.57**	19.12 ± 2.73**	19.75 ± 4.00**	35.88 ± 10.36
			Females		
0	2.27 ± 0.65	29.08 ± 3.43	46.34 ± 6.86	60.01 ± 9.21	71.38 ± 12.63
5	-0.46 ± 0.61	18.71 ± 2.55	31.71 ± 2.49	37.29 ± 3.69*	--
20	-1.30 ± 0.50**	14.80 ± 3.22*	22.53 ± 3.55**	21.84 ± 3.40**	--
50	-2.31 ± 1.00**	7.59 ± 3.82**	14.63 ± 4.43**	17.11 ± 4.12**	25.96 ± 7.42*

[a]Based on eight controls and eight high-dose animals/sex and five low- and mid-dose animals/sex.

[b]Percent body weight gain calculated as: $\frac{\text{Current Body Weight (bw)} - \text{Day 1 bw}}{\text{Day 1 bw}} \times 100.$

[c]Based on three control and three high-dose recovery animals/sex held for 4 additional weeks.

*Significantly different from control at $p < 0.05$.

**Significantly different from control at $p < 0.01$.

weight gains of high-dose males indicated a recovery of 0.86 kg from weeks 53 to 57 compared to a body weight loss of 0.3 kg for concurrent controls during this same time.

Body weights of high-dose females were significantly depressed from study weeks 6 to 9 ($p < 0.05$), and during weeks 11 to 53 ($p < 0.01$). During study weeks 4, 5, and 10, body weights of these animals were slightly but not significantly depressed. Body weights of mid-dose females were significantly depressed from study weeks 22 to 43 ($p < 0.05$) and from weeks 44 to 53 ($p < 0.01$). Percent weight gains of dosed females were depressed throughout the study; weight gains of mid- and high-dose animals were depressed ($p < 0.01$) 52 to 80% and 67 to 90%, respectively, during study weeks 0-1 to 0-3, 0-11 to 0-12, and 0-22 to 0-53. During weeks 0-4 to 0-9 and 0-13 to 0-21, percent weight gains of mid- and high-dose females were depressed 49 to 62% ($p < 0.05$) and 73 to 89% ($p < 0.01$), respectively. Low-dose females exhibited significantly reduced weight gains during weeks 28 to 53; however, these changes were not significantly different from controls at any interval of analysis. Body weight gains of high-dose females indicated a recovery of 0.6 kg from weeks 53 to 57. The body weights and body weight gains of females appeared to be depressed to a greater extent when compared to those of males.

Because of poor physical condition, one high-dose male (animal No. 734) was administered powdered diet or canned dog food on two days during the latter phase of the study. The dog was returned to the pelleted diet as the physical condition improved.

3. Food Consumption and Compound Intake: Food consumption was determined weekly.

Results: Representative results of mean food consumption are summarized in Table 4. The food consumption of high-dose males was significantly depressed ($p < 0.05$, $p < 0.01$) throughout the dosing period when compared to concurrent controls. The food consumption of one high-dose male (animal No. 734) was severely reduced from study weeks 45 to 47. During recovery, the food consumption of high-dose males was similar to concurrent controls, and the food consumption of low- and mid-dose males during the dosing period was comparable to control consumption. The food consumption of high-dose females was nonsignificantly depressed throughout the dosing period; depressions were significant ($p < 0.05$, $p < 0.01$) at weeks 1, 7, 17-20, and 39. The food consumption of these animals during recovery, and of low- and mid-dose animals during dosing, was similar to that of concurrent controls.

TABLE 4. Representative Results of Mean Food Consumption for Dogs Administered Prometon for 53 Weeks[a]

Dose Level (mg/kg/day)	Food Consumption (g/dog/week ± SE) at Weeks: 1	13	26	53	56[b]
			Males		
0	389.4 ± 7.78	388.3 ± 7.73	387.5 ± 9.01	348.3 ± 16.78	368.0 ± 29.05
5	370.8 ± 16.21	388.0 ± 12.00	381.4 ± 11.78	360.2 ± 15.79	--
20	354.2 ± 21.56	393.8 ± 6.20	378.2 ± 21.80	353.0 ± 19.13	--
50	323.0 ± 16.21**	322.5 ± 11.64**	313.1 ± 14.96**	322.0 ± 16.20	395.7 ± 4.33
			Females		
0	368.1 ± 21.75	368.4 ± 14.10	361.9 ± 20.76	304.9 ± 20.54	358.0 ± 36.50
5	312.4 ± 20.55	343.2 ± 8.32	328.0 ± 32.93	278.0 ± 7.29	--
20	291.8 ± 19.61*	339.6 ± 19.09	351.0 ± 21.13	345.6 ± 24.40	--
50	304.5 ± 25.07*	325.6 ± 19.47	318.5 ± 7.51	301.3 ± 23.06	386.0 ± 14.00

[a]Based on eight controls and eight high-dose animals/sex and five mid- and low-dose animals/sex.

[b]Based on three control and three high-dose recovery animals/sex held for 4 additional weeks.

*Significantly different from control at $p < 0.05$.

**Significantly different from control at $p < 0.01$.

4. Ophthalmology: Ophthalmological examinations were performed prior to dosing and at 3, 6, 9, 12, and 13 months using focal illumination and indirect ophthalmology.

Results: The incidence of mydriasis (8/8 in males and females) and ptosis (4/8 in males and 7/8 in females) was increased in high-dose males and females; these findings were considered by the reviewer to be related to dosing. Other ocular findings were sporadic and were considered to be incidental.

5. Hematology and Clinical Chemistry: Blood was collected by venous puncture prior to study initiation and at weeks 14, 27, and 53 for hematology and clinical analysis from all dosed animals. Blood was collected from recovery animals at week 57, and blood was collected from male No. 734 at study weeks 46 and 47 for additional analyses. Animals were fasted overnight prior to bleeding. The CHECKED (X) parameters were examined:

a. Hematology:

X Hematocrit (HCT)†
X Hemoglobin (HGB)†
X Leukocyte count (WBC)†
X Erythrocyte count (RBC)†
X Platelet count†
X Reticulocyte count (RETIC)
Red cell morphology
X Heinz bodies
X Leukocyte differential count
X Mean corpuscular HGB (MCH)
X Mean corpuscular HGB concentration (MCHC)
X Mean corpuscular volume (MCV)
X Coagulation:prothrombin time (PT)
X Methemoglobin[a]

Results: There were no hematological changes which were considered to be related to dosing. Erythrocyte indices (hemoglobin, hematocrit, erythrocyte counts, and calculated indices) of mid-dose and high-dose males and high-dose females were slightly depressed sporadically throughout the study; however, these changes were slight (within 7% difference from concurrent controls), and were not considered to be of toxicological importance. In addition, baseline measurements of many parameters indicated depressed values prior to dosing (e.g., the hemoglobin concentration of high-dose males was depressed 8% when compared to concurrent controls prior to dosing and 6.5% and 9% at weeks 26 and 52, respectively). Reticulocyte counts of mid- and high-dose males were depressed 40 and

---

[a]Methemoglobin was not measured prior to dosing.

†Recommended by Subdivision F (October 1984) Guidelines.

37%, respectively, at week 26, and 36 and 26%, respectively, at week 52 as compared to respective baseline depressions of 27 and 7% for these mid- and high-dose animals. Reticulocyte counts of dosed females were similar to concurrent controls. Lymphocyte and neutrophil values of dosed males and females varied slightly at weeks 26 and 52; changes between sexes were inconsistent and are not considered to be of any toxicological significance.

b. Clinical Chemistry:

	Electrolytes
X	Calcium†
X	Chloride†
	Magnesium
X	Phosphorus†
X	Potassium†
X	Sodium†

	Enzymes
X	Alkaline phosphatase (ALP)
	Cholinesterase
X	Creatine phosphokinase†
	Lactic acid dehydrogenase
X	Serum alanine aminotransferase (SGPT)†
X	Serum aspartate aminotransferase (SGOT)†
X	Gamma glutamyltransferase (GGT)

	Other
X	Albumin†
X	Albumin/globulin ratio
X	Blood creatinine†
X	Blood urea nitrogen†
X	Cholesterol†
X	Globulins
X	Glucose†
X	Total bilirubin†
	Direct bilirubin
X	Total protein†
	Triglycerides

Results: Table 5 summarizes mean clinical chemistry data. Mean total protein, globulin, and cholesterol levels of high-dose males were significantly ($p < 0.05$, $p < 0.01$) decreased when compared to concurrent controls at study weeks 13, 26, and 52; total protein levels were decreased by 10 to 14%, globulin was depressed by 18 to 24%, and cholesterol was depressed by 22 to 26% in these animals. Albumin levels of mid- and high-dose males were slightly but significantly ($p < 0.05$) depressed from 9 to 11% at study week 26 only. There were no comparable changes in the clinical chemistry parameters of dosed females. Creatinine phosphokinase levels were slightly but significantly ($p < 0.01$) depressed in high-dose males at 13 weeks and in mid- and high-dose males at 26 and 52 weeks.

---

†Recommended by Subdivision F (November 1984) Guidelines.

TABLE 5. Representative Clinical Chemistry Results (± SE) for Dogs Administered Prometon for 53 Weeks[a]

	Dose Level (mg/kg/day)							
	Males				Females			
Parameter/Week	0	5	20	50	0	5	20	50
Total Protein (g/dL)								
Baseline	6.01 ± 0.10	6.52 ± 0.33	6.18 ± 0.10	6.39 ± 0.11	5.43 ± 0.08	5.72 ± 0.13	5.66 ± 0.06	5.76 ± 0.09
13	5.80 ± 0.09	5.74 ± 0.12	5.98 ± 0.09	5.50 ± 0.10*	5.59 ± 0.08	5.78 ± 0.20	5.68 ± 0.12	5.50 ± 0.15
26	6.20 ± 0.12	6.02 ± 0.17	6.08 ± 0.10	5.63 ± 0.08**	6.03 ± 0.09	6.04 ± 0.24	5.82 ± 0.06	5.85 ± 0.18
52	6.43 ± 0.17	6.00 ± 0.21	6.08 ± 0.12	5.73 ± 0.10**	6.14 ± 0.08	6.10 ± 0.16	6.02 ± 0.14	5.81 ± 0.17
Albumin (g/dL)								
Baseline	2.93 ± 0.07	3.08 ± 0.12	3.18 ± 0.07	3.33 ± 0.08	2.80 ± 0.05	3.00 ± 0.07	2.82 ± 0.09	2.86 ± 0.06
13	3.10 ± 0.05	3.18 ± 0.06	3.10 ± 0.07	3.08 ± 0.06	3.13 ± 0.06	3.24 ± 0.08	3.24 ± 0.02	3.09 ± 0.07
26	3.18 ± 0.05	3.10 ± 0.06	2.92 ± 0.07*	2.99 ± 0.04*	3.15 ± 0.04	3.18 ± 0.04	3.32 ± 0.04	3.21 ± 0.09
52	3.25 ± 0.09	3.28 ± 0.06	3.18 ± 0.02	3.20 ± 0.06	3.21 ± 0.07	3.38 ± 0.07	3.32 ± 0.05	3.15 ± 0.07
Globulin (g/dL)								
Baseline	3.09 ± 0.08	3.44 ± 0.24	3.00 ± 0.07	3.06 ± 0.11	2.63 ± 0.07	2.72 ± 0.12	2.84 ± 0.08	2.90 ± 0.07
13	2.70 ± 0.07	2.56 ± 0.07	2.88 ± 0.07	2.43 ± 0.07*	2.46 ± 0.06	2.54 ± 0.15	2.44 ± 0.13	2.41 ± 0.11
26[b]	3.03 ± 0.08	2.92 ± 0.16	3.16 ± 0.10	2.64 ± 0.07**	2.88 ± 0.10	2.86 ± 0.22	2.50 ± 0.10	2.64 ± 0.36
52	3.18 ± 0.15	2.72 ± 0.20	2.92 ± 0.13	2.53 ± 0.09**	2.68 ± 0.74	2.72 ± 0.23	2.70 ± 0.28	2.66 ± 0.42
Cholesterol (mg/dL)								
Baseline	144.0 ± 8.79	146.2 ± 15.33	142.8 ± 5.89	136.3 ± 4.39	127.9 ± 5.49	131.2 ± 8.74	133.0 ± 4.34	136.1 ± 12.07
13	151.1 ± 10.77	148.4 ± 14.07	147.8 ± 9.08	119.6 ± 4.60*	154.6 ± 8.06	135.0 ± 9.10	156.0 ± 16.01	128.4 ± 10.59
26	145.6 ± 12.28	124.4 ± 11.83	134.4 ± 7.66	117.9 ± 4.03*	182.9 ± 15.89	179.8 ± 25.29	141.6 ± 18.36	152.6 ± 19.20
52	126.8 ± 10.61	119.8 ± 15.46	119.8 ± 10.69	101.5 ± 5.72	166.5 ± 11.68	160.6 ± 12.87	129.6 ± 24.30	154.0 ± 25.06

[a]Based on eight controls and eight high-dose animals/sex and five mid- and low-dose animals/sex.

[b]Globulin summary data for females recalculated by the reviewers for weeks 26 and 52 owing to inability to read the summary tables.

*Significantly different from control at $p < 0.05$.

**Significantly different from control at $p < 0.01$.

Creatinine phosphokinase was slightly increased in mid- and high-dose females at weeks 26 and 52 when compared to concurrent controls. These changes in creatinine phosphokinase were not considered to be of toxicological importance.

6. Urinalysis: Urine was collected from fasted males at study initiation and weeks 14, 27, and 53 (recovery animals) by catheterization and from sacrificed animals at weeks 53 and 57 by bladder puncture. Urine was collected from fasted females at weeks 53 and 57 by bladder puncture. 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

Results: There were no compound-related changes in urinalyses parameters.

7. Sacrifice and Pathology: All animals that were sacrificed on schedule were subject to gross pathological examination, and the CHECKED (X) tissues were collected for histological examination. In addition, the (XX) organs were weighed:

---

†Recommended by Subdivision F (October 1984) Guidelines.

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

Results:

a. Organ Weights: There were no effects of dosing on organ weights. Slight changes in absolute or relative weights of adrenals and kidneys in males, and of spleen, pituitary, kidneys, and adrenals in females were not consistent between males and females and were not dose-related; there were no correlating clinical chemistry or histological changes.

b. Gross Pathology: Gross lesions were infrequent and sporadic, and were not considered to be related to dosing.

---

†Recommended by Subdivision F (October 1984) Guidelines.

c. Microscopic Pathology:

1) Nonneoplastic: There were no compound-related nonneoplastic lesions. Two high-dose males (animal Nos. 7397 and 1734) exhibited testicular atrophy; these and other minor, sporadically exhibited cysts or inflammation were not considered to be a result of dosing.

2) Neoplastic: There were no neoplastic lesions.

D. STUDY AUTHORS' CONCLUSIONS:

The 53-week administration of prometon to male and female beagle dogs at dose levels of 0, 5, 20, or 50 mg/kg/day resulted in a high incidence of emesis and lethargy at the two highest doses. Diarrhea, ptosis, mydriasis, and tremors also occurred at 50 mg/kg. Body weights and food consumption of mid- and high-dose animals were depressed when compared to concurrent controls. Slight changes in hematological and clinical chemistry parameters were not indicative of target organ toxicity. There were no changes in organ weights or gross or histologic changes which were considered to be related to dosing. All effects were reversible following the 4-week recovery period. The LOEL is 20 mg/kg/day and the NOEL is 5 mg/kg/day. The maximum tolerated dose is 50 mg/kg/day due to decreased body weights.

E. REVIEWERS' DISCUSSION AND INTERPRETATION OF RESULTS:

The study design was adequate, and the conduct of the study was acceptable. However, technical errors in dosing were reported for one low-dose male, one high-dose male, and five mid-dose females; the study authors indicated that these dosing errors had no adverse effect on the study.

We agree with the study authors that prometon induced significant effects on body weights and on the incidence of emesis, diarrhea, lethargy, mydriasis, and ptosis in mid- and high-dose animals. Based on clinical observations and the depression of body weight, the LOEL is 20 mg/kg/day, and the NOEL is 5 mg/kg/day.