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

JUL 10 1997

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM

Subject: BCDMH. Review of Toxicology Data.
DP Barcode: D229526 Submission No.: S511040
Tox. Chem. No.: 114A PC Code No.: 006315-005785

To: Walt Waldrop
Reregistration Branch III
Special Review and Reregistration Division (7508W)

From: Raymond K. Locke, Toxicologist Raymond K. Locke 7/9/97
Reregistration Branch I
Health Effects Division (7509C)

Thru: Whang Phang, Ph.D., Branch Senior Scientist
Reregistration Branch I
Health Effects Division (7509C) 7/10/97

Registrant: Great Lakes Chemical Corporation
P.O. Box 2200
West Lafayette, IN

Action Requested: Review toxicology data (MRID 44095901) submitted to support reregistration of BCDMH and indicate whether these data meet the requirements for a chronic toxicity/oncogenicity toxicity study in rats (Guideline 83-5).

Conclusion: This study was adequately conducted and supports the reregistration of BCDMH. The data presented demonstrate that, under the study conditions, the study may be classified as follows:

In a chronic/oncogenicity toxicity study (MRID 44095901), dimethylhydantoin (DMH) (97.3%, 97.1% and 93.5% a.i. for lot # 6, # 2412-67-D1, and # 2412-67-D2, respectively) was administered to 80 Crl:CD®BR rats/sex/dose in the diet at dose levels of 0, 100, 320, or 1000 mg/kg/day for 104-105 weeks. The same doses of DMH were given to 20 rats/sex/dose in a 52-week interim study. The concentration of DMH in the diets was adjusted weekly according to body weight and food consumption measurements to achieve the target doses.

No treatment-related statistically significant increases in the incidences of any lesion were seen at the termination of this 105-week study. However, the animals that died during the study tended

to die earlier at the high dose. Mortality during treatment weeks 52-79 was 11% in control males and 15% in high dose males; 7% in control females, and 27% ($p \leq 0.001$) in high dose females. Significantly increased incidences of enlarged pituitary glands were found in high dose early decedent males (control, 20%; high dose, 38%, $p \leq 0.05$) and females (control, 33%; high dose, 61%, $p \leq 0.05$). Pituitary pars distalis adenomas were the most common cause of early deaths in the study and contributed to the increased mortality seen during weeks 52-79 at the high dose. Increased incidences of mammary galactoceles occurred in high dose early decedent females (control, 31%; high dose, 59%, $p \leq 0.01$). The incidence of testicular fibroid vascular degeneration was increased in high dose males (control, 8%; high dose, 27%, $p \leq 0.01$). The incidence of iliac lymph node lymphangiectasia was increased in high dose males (control, 13%; high dose, 26%) and females (control, 1%; high dose, 9%). The number of times that dried yellow matting in the urogenital area was observed over the 24-month treatment period increased about 3-fold in high dose males and females compared to controls. Urine volume roughly doubled in high dose males and females, and specific gravity decreased during week 77. Kidney tubule dilation was increased in high dose males at 52 weeks (control, 65%; high dose, 100%, $p \leq 0.01$). Lung mineralization at 52 weeks was increased in males at 1000 mg/kg/day (68%, $p \leq 0.05$) compared to the controls (30%). The mean absolute brain weight was slightly decreased in high dose males (4%, $p \leq 0.05$) compared to the controls at 52 weeks.

The LOEL is 1000 mg/kg/day for males and females, based on enlarged pituitary glands in early decedents of both sexes; increased mortality earlier in the study, especially in females; increased mammary galactoceles in early decedents of both sexes; and testicular fibrinoid vascular degeneration in early decedent males. The NOEL for both sexes is 320 mg/kg/day.

At the doses tested, there <u>was not</u> a treatment related increase in tumor incidence after 105 weeks of treatment with DMH. The increase at 52 weeks in the incidences of pituitary adenomas in the pars distalis in high dose females (control, 10%; high dose, 50%, $p \leq 0.01$) and males (control, 10%; high dose 32%, N.S.) was not dose-related. There were no differences in pituitary adenoma incidences between control and high dose groups by the end of the 105-week study. The high dose of 1000 mg/kg/day meets the limit dose requirement for non-toxic agents.

This oncogenicity study in the rat is acceptable, and does satisfy the guideline requirement for a chronic/oncogenicity study (83-5) in rats.

Classification: Acceptable/Guideline

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DIMETHYLHYDANTOIN **Chronic Oral/Oncogenicity Study (83-5)**

EPA Reviewer: W. Greear, M.P.H., D.A.B.T. W. Greear, Date 6/3/97
Review Section IV, Toxicology Branch I (7509C)
EPA Secondary Reviewer:
M. Copley, D.V.M, D.A.B.T. Myron Copley, Date 7/3/97
Toxicology Branch I (7509C)

DATA EVALUATION RECORD

STUDY TYPE: Combined Chronic/Oncogenicity Feeding - Rat
OPPTS 870.4300 [§83-5]

DP BARCODE: D229526
P.C. CODE: 006315
SUBMISSION CODE: S511040
TOX. CHEM. NO.: 114A

TEST MATERIAL (PURITY): Dimethylhydantoin (Lot # 6, 97.3%; Lot # 2412-67-D1, 97.1%; Lot # 2412-67-D2, 93.5%)

SYNONYMS: DMH

CITATION: Naas, D. (1996) Combined 24-month toxicity/oncogenicity study in rats with DMH. WIL Research Laboratories, Inc., Ashland, Ohio. Laboratory study number WIL-12258, July 30, 1996. MRID 44095901. Unpublished.

SPONSOR: Great Lakes Chemical Corporation, West Lafayette, Indiana.

EXECUTIVE SUMMARY: In a chronic/oncogenicity toxicity study (MRID 44095901), dimethylhydantoin (DMH) (97.3%, 97.1% and 93.5% a.i. for lot # 6, # 2412-67-D1, and # 2412-67-D2, respectively) was administered to 80 Crl:CD®BR rats/sex/dose in the diet at dose levels of 0, 100, 320, or 1000 mg/kg/day for 104-105 weeks. The same doses of DMH were given to 20 rats/sex/dose in a 52-week interim study. The concentration of DMH in the diets was adjusted weekly according to body weight and food consumption measurements to achieve the target doses.

No treatment-related statistically significant increases in the incidences of any lesion were seen at the termination of this 105-week study. However, the animals that died during the study tended to die earlier at the high dose. Mortality during treatment weeks 52-79 was 11% in control males and 15% in high dose males; 7% in control females, and 27% ($p \leq 0.001$) in high dose females. Significantly increased incidences of enlarged pituitary glands were found in high dose early decedent males (control, 20%; high dose, 38%, $p \leq 0.05$) and females (control, 33%; high dose, 61%, $p \leq 0.05$). Pituitary pars distalis adenomas were the most common cause of early deaths in the study and contributed to the increased mortality seen during weeks 52-79 at the high dose. Increased incidences of mammary galactoceles occurred in high dose early decedent females (control, 31%; high

dose, 59%, $p \leq 0.01$). The incidence of testicular fibroid vascular degeneration was increased in high dose males (control, 8%; high dose, 27%, $p \leq 0.01$). The incidence of iliac lymph node lymphangiectasia was increased in high dose males (control, 13%; high dose, 26%) and females (control, 1%; high dose, 9%). The number of times that dried yellow matting in the urogenital area was observed over the 24-month treatment period increased about 3-fold in high dose males and females compared to controls. Urine volume roughly doubled in high dose males and females, and specific gravity decreased during week 77. Kidney tubule dilation was increased in high dose males at 52 weeks (control, 65%; high dose, 100%, $p \leq 0.01$). Lung mineralization at 52 weeks was increased in males at 1000 mg/kg/day (68%, $p \leq 0.05$) compared to the controls (30%). The mean absolute brain weight was slightly decreased in high dose males (4%, $p \leq 0.05$) compared to the controls at 52 weeks.

The LOEL is 1000 mg/kg/day for males and females, based on enlarged pituitary glands in early decedents of both sexes; increased mortality earlier in the study, especially in females; increased mammary galactoceles in early decedents of both sexes; and testicular fibrinoid vascular degeneration in early decedent males. The NOEL for both sexes is 320 mg/kg/day.

At the doses tested, there <u>was not</u> a treatment related increase in tumor incidence after 105 weeks of treatment with DMH. The increase at 52 weeks in the incidences of pituitary adenomas in the pars distalis in high dose females (control, 10%; high dose, 50%, $p \leq 0.01$) and males (control, 10%; high dose 32%, N.S.) was not dose-related. There were no differences in pituitary adenoma incidences between control and high dose groups by the end of the 105-week study. The high dose of 1000 mg/kg/day meets the limit dose requirement for non-toxic agents.

This oncogenicity study in the rat is acceptable, and does satisfy the guideline requirement for a chronic/oncogenicity study (83-5) in rats.
<u>COMPLIANCE</u>: Signed and dated GLP, Quality Assurance, Data Confidentiality, and Flagging statements were provided.

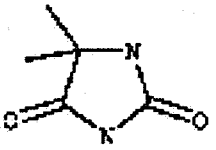
I. MATERIALS AND METHODS

A. <u>MATERIALS</u>

1. <u>Test material</u>: Dimethylhydantoin (DMH)

   Description: white crystalline solid
   Lot/Batch #: 6 (used weeks 0 to 25), # 2412-67-D1 (used weeks 25 to 89), # 2412-67-D2 (used weeks 89 through study termination)
   Purity: 97.3%, 97.1%, 93.5% a.i. for lot #'s 6, 2412-67-D1, and 2412-67-D2, respectively
   Stability of compound: Stable at room temperature.
   CAS #: 77-71-4

Structure:



2. Vehicle and/or positive control

Test substance was mixed with food.

3. Test animals: Species: rat

Strain: Crl:CD®BR
Age and weight at study initiation: males and females, 6 weeks; weights males, 141-191 g; females, 116-157 g.
Source: Charles River Breeding Laboratories, Portage, Michigan.
Housing: rats were housed individually in wire-mesh cages suspended above cage-board. The cage-boards were changed three times per week.
Diet: Purina Certified Rodent Chow® #5002 ad libitum
Water: filtered tap water ad libitum
Environmental conditions:
Temperature: set at 72 ± 4° F; actual range: 61-79° F.
Humidity: set at 30-70%; actual range: 30-91%.
Air changes: not supplied
Photoperiod: 12 hours light/12 hours dark
Acclimation period: 12 days

B. STUDY DESIGN

1. In life dates

Start: Sept. 22, 1992; end: Sept. 30, 1994.

2. Animal assignment

Animals were assigned randomly within weight limitations to the test groups in Table 1.

TABLE 1: Study design						
Test Group	Dose to Animal[a] mg/kg/day		Main Study 24 Months No. Animals		Interim Sac. 12 Months No. Animals	
	Male	Female	Male	Female	Male	Female
Control	0	0	80	80	20	20
Low	100 (101)	100 (101)	80	80	20	20
Mid	320 (323)	320 (323)	80	80	20	20
High	1000 (1008)	1000 (1014)	80	80	20	20

Taken from pp. 27 and 43, MRID 44095901.
[a]Target dose (calculated compound consumption). Dietary concentration was adjusted weekly to maintain stated dose levels.

3. Dose selection

The dose selections were based on the results from previous studies, descriptions of which were not provided..

4. Diet preparation and analysis

Diets were prepared weekly by mixing appropriate amounts of test substance with Purina Certified Rodent Chow® #5002. In preparing the dietary mixtures and dose calculations, the test substance was assumed to be 100% DMH. Dietary concentrations of DMH were adjusted for body weights and food intake to achieve the target doses. The DMH was ground through a Wiley mill with a #30 mesh screen and weighed. A premix was prepared by mixing the weighed amount of DMH with 5 kg rodent feed in a Hobart mixer. The dietary mixture for each of the treated groups was prepared by diluting the premix with a predetermined amount of food and mixing in a V-twin shell mixer for 15 minutes. Separate batches of the mixtures were prepared for males and females at each dose level. The diets were then stored at room temperature. Samples from each dose level were taken and checked for the stability of DMH at 7 and 14 days by comparing the concentrations in frozen samples to the

concentrations of DMH in samples stored at room temperature. The homogeneity of all 3 dietary mixtures was tested prior to the initiation of dosing by analyzing samples taken from the top, middle, and bottom of the storage container for each dose level. Dietary samples taken from the middle of the container for each dose level during study weeks 0, 1, 2, 3, 7, 11, 24, 37, 50, 63, 76, 89, and 102 were analyzed to check the DMH concentrations. The analyses were performed utilizing gas chromatography with flame ionization detection.

Results -
Homogeneity Analysis: Analysis of samples taken from the top, middle, and bottom of each dietary mixture, a total of 12 samples for each dose level, resulted in mean concentrations that were 113-123% of the target low dose, 102-113% of the target mid dose, and 104-109% of the target high dose. The resulting coefficients of variation were 4.2%, 5.9%, and 2.5% for the low, mid, and high doses, respectively.

Stability Analysis: The concentrations of DMH in the 7-day frozen samples were 0.830 ± 0.023, 2.78 ± 0.057, and 9.07 ± 0.23 g/kg for the low, mid, and high doses, respectively. The samples stored at room temperature for 7 days resulted in concentrations of 0.819 ± 0.021, 2.70 ± 0.043, and 8.38 ± 0.17 g/kg. Analysis of samples stored for 14 days gave concentrations of 0.980 ± 0.011, 3.05 ± 0.030, and 9.60 ± 0.032 g/kg for the frozen samples; and 0.955 ± 0.015, 2.92 ± 0.019, and 8.97 ± 0.11 g/kg for the low, mid and high doses, respectively, stored at room temperature. Although the concentrations of samples stored at room temperature were slightly, but consistently, lower than the frozen samples, all samples were within ± 15% of the target dose. The differences, in most cases, were also within the error of the determinations. The stored DMH stock was analyzed at approximately 6-month intervals and at study termination. The 1-year analysis was omitted. The samples taken at 18 months gave a mean DMH purity of 94.9%, but 2 of the 5 samples appeared to be non-representative. Eliminating the 2 outlying samples gave a purity of 96.5%. The purity of lot # 2412-67-D2 used at study termination was 93.5%.

Concentration Analysis: The overall means of the samples taken throughout the study as percent of target dose for the low, mid, and high doses, respectively are: 111%, 109%, and 109% for males, and 111%, 109%, and 106% for females. Five individual samples were found to be 116% of the target dose, one sample was 117%, one was 118%, and one was 120%. All

other samples were within ± 15% of the target concentrations.

The analytical data indicated that the mixing procedure was adequate and that the variance between nominal and actual dosage to the animals was acceptable.

5. Statistics

The analyses were conducted using two-tailed tests for significance levels of $p \leq 0.05$ and $p \leq 0.01$, comparing the treated groups to the control group by sex. All means were presented with standard deviations and the numbers of sampling units used to calculate the means. Body weight, body weight change, food consumption, clinical pathologic data and absolute and relative organ weight data were subjected to a one-way analysis of variance, followed by Dunnett's Test. Clinical laboratory values for cell types that occurred at a low incidence were not subjected to statistical analysis.

Fisher's Exact Test was used to compare survival data between the control and treated groups for the chronic and oncogenicity subgroups. In addition, Fisher's Exact Test was used to compare the incidence of testicular atrophy in the treated groups with the control group.

Fisher's Exact Test and the Chi-square Test were not used by the study authors to compare the tumor incidence between the control and treated groups as no remarkable differences in tumor incidence were apparent. Fisher's Exact Test was applied by the reviewer to compare selected tumor incidences. Results were tabulated as significant at $p \leq 0.05$.

C. METHODS

1. Observations

All animals were inspected twice a day for mortality and morbidity, and daily for clinical signs. Each animal was palpated for masses and given a detailed physical examination weekly beginning one week prior to the initiation of dosing.

2. Body weight

Animals were weighed weekly throughout the study beginning one week prior to study initiation.

3. Food consumption and compound intake

Food consumption was measured weekly beginning one week prior to the study initiation. Weekly averages were reported and food intake was calculated as g/animal/day. The concentration of DMH in the diet was adjusted to achieve the target mg/kg/day doses. Food efficiency (body weight gain in kg/food consumption in kg per unit time X 100) values were not calculated by the study authors.

4. Ophthalmoscopic examination

The eyes of all main study animals were examined with an indirect ophthalmoscope prior to study initiation, during week 51, and during week 103. Eyes were dilated prior to the examinations.

5. Blood was collected for hematology and clinical chemistry analysis from the tail vein of fasted animals during study weeks 11, 24, and 50 of the 12-month interim groups, and during weeks 12, 25, 51, 77 and 105 for the 24-month oncogenicity groups. Blood was taken from the vena cava of surviving animals under carbon dioxide anesthesia at study termination. Hematology evaluations were performed on 10 rats/sex/group in the 12-month interim groups and on 20 rats/sex/group in the 24-month oncogenicity groups. Clinical chemistry evaluations were done on 10 animals/sex/group in both the 12-month and 24-month groups. The clinical chemistry evaluations were done on different animals than those selected for hematology except for week 105 at study termination. The CHECKED (X) parameters were examined.

a. Hematology

X		X	
X	Hematocrit (HCT)	X	Leukocyte differential
X	Hemoglobin (HGB)	X	count*
X	Leukocyte count (WBC)	X	Mean corpuscular HGB (MCH)
X	Erythrocyte count (RBC)	X	Mean corpusc. HGB
X	Platelet count		conc.(MCHC)
	Blood clotting measurements	X	Mean corpusc. volume (MCV)
	(Thromboplastin time)		Reticulocyte count
	(Thromboplastin time)		Erythrocyte morphology
	(Clotting time)		
	(Prothrombin time)		

*Minimum required for carcinogenicity studies (only on control and high dose groups unless effects are observed based on Subdivision F Guidelines).

b. Clinical chemistry

X	ELECTROLYTES	X	OTHER
X	Calcium	X	Albumin
X	Chloride	X	Blood creatinine
	Magnesium	X	Blood urea nitrogen
X	Phosphorus	X	Total Cholesterol
X	Potassium	X	Globulins
X	Sodium	X	Glucose
		X	Total bilirubin
		X	Total serum protein (TP)
			Triglycerides
X	ENZYMES		Serum protein
	Alkaline phosphatase (ALK)	X	electrophoresis
	Cholinesterase (ChE)		A/G ratio
	Creatine phosphokinase		
X	Lactic acid dehydrogenase (LDH)		
X	Serum alanine amino-transferase		
X	(also SGPT)		
	Serum aspartate amino-transferase (also SGOT)		
	Gamma glutamyl transferase (GGT)		
–	Glutamate dehydrogenase		

6. Urinalysis

Urine was collected overnight for analysis using metabolism cages from 10 animals per group in the 12-month interim study during treatment weeks 11, 24, and 50, and from 10 animals per group in the 24-month oncogenicity study during weeks 12, 25, 51, 77, and 104. Urine was collected from the same animals used for serum clinical chemistry evaluations. The CHECKED (X) parameters were evaluated.

X	Appearance*	X	Glucose*
X	Volume*	X	Ketones*
	Specific gravity*		Bilirubin*
X	pH	X	Blood*
	Sediment		Nitrites
X	(microscopic)*	X	Urobilinogen
	Protein		
X		X	
X		X	
X		X	

*Required for chronic studies.

7. Sacrifice and pathology

All animals that died spontaneously and those sacrificed by exsanguination under CO_2 anesthesia before or at the scheduled termination of the 12- or 24-month studies were subjected to gross pathological examination. The CHECKED (X) tissues were collected and preserved in 10% neutral buffered formalin for histological examination. The (XX) organs from all animals at scheduled necropsies, in addition, were weighed.

X	DIGESTIVE SYSTEM	X	CARDIOVASC./HEMAT.	X	NEUROLOGIC
	Tongue	X	Aorta*	XX	Brain*+
	Salivary	XX	Heart*	X	Periph.nerve*
X	glands*	X	Bone marrow*	X	Spinal cord (3 levels)*
X	Esophagus*	X	Lymph nodes*		
X	Stomach*	X	Spleen*	X	Pituitary*
X	Duodenum*	X	Thymus*	X	Eyes (optic n.)*
X	Jejunum*				
X	Ileum*		UROGENITAL		GLANDULAR
X	Cecum*	XX	Kidneys*+	XX	Adrenal gland*
X	Colon*	X	Urinary		Lacrimal
X	Rectum*	XX	bladder*	X	(Harder's) gland
X	Liver*+	X	Testes*+	X	Mammary gland*
	Gall bladder*	X	Epididymides	X	Parathyroids*
X	Pancreas*	XX	Prostate	X	Thyroids*
		X	Seminal vesicle		Preputial gland
	RESPIRATORY	X	Ovaries*+		
X	Trachea*		Uterus*	X	OTHER
	Lung*		Vagina	X	Bone*
	Nose			X	Skeletal muscle*
	Pharynx				Skin*
	Larynx				All gross lesions and masses*

* Required for carcinogenicity studies based on Subdivision F Guidelines.
+ Organ weight required in chronic studies.

II. RESULTS

A. OBSERVATIONS

1. Toxicity

Clinical signs seen in daily exams that were different between treated animals and controls are

shown in Table 2. The incidence of hypoactivity in males was slightly, but not significantly, increased at the high dose (control, 34%; high dose, 43%), and the number of times it was observed was increased at the mid and high doses (by 31% and 52%, respectively) compared to the control group. Incidences of head tilt (control, 5%; high dose, 18%) and impaired equilibrium (control, 6%; high dose, 16%,) were significantly ($p \leq 0.05$) increased in high dose males. The incidences of head tilt and impaired equilibrium were higher in the female control group than the male control group. The incidence of hypoactivity was slightly higher in high dose females (control 39%; high dose, 44%), but the incidence of head tilt in high dose females was the same as the female control group. However, the frequencies of hypoactivity and head tilt were increased in females at the high dose (by 15% and 51%, respectively, compared to controls). One of the most commonly observed findings in high dose animals, especially as the rats aged, was the appearance of yellow matting in the urogenital areas of both sexes. Males were affected more than females. The number of high dose males with dried yellow matting in the urogenital area was increased by ~91% over the controls ($p \leq 0.001$); females were increased by ~33% at the high dose.

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TABLE 2: Clinical observations in the 24-month study with DMH.

Clinical Observation	Total no. of times clinical observations were made over 2 yrs./No. Rats with finding/percent[a]			
	Control	100 mg/kg/day	320 mg/kg/day	1000 mg/kg/day
	MALES			
Hypoactive	216/27/34%	209/20/25%	284/27/34%	329/34/43%
Head tilt	11/4/5%	59/8/10%	53/6/8%	85/14*/18%
Impaired equilibrium	13/5/6%	21/7/9%	9/2/3%	25/13*/16%
Dried yellow matting, urogenital area	281/22/28%	160/25/31%	458/34*/43%	933/42***/53%
_	**FEMALES**			
Hypoactive	331/31/39%	419/34/43%	437/32/40%	381/35/44%
Head tilt	158/19/24%	201/22/28%	294/24/30%	239/19/24%
Impaired equilibrium	53/15/19%	39/12/15%	47/14/18%	35/10/13%
Dried yellow matting, urogenital area	168/24/30%	280/27/34%	339/31/39%	447/32/40%

Data extracted from Table 3, pp. 125-135, MRID 44095901
*p ≤ 0.05, ***p ≤ 0.001, significantly different from controls.
Fisher exact test performed by the reviewer.
[a]Percentage was based on 80 rats/group.

2. Mortality

The cumulative mortality at various times during the treatment period for each dose level and the overall percent survival are given in Table 3. The mortality was slightly increased at the high dose in both sexes compared to the control group (~8% in males and 22% in females); however, the increases were not statistically significant (Fisher's Exact Test) and

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the mortality was not dose-related. However, the number of high dose female early decedents from week 52-79 was significantly ($p \leq 0.001$) greater than the number of controls dying over the same time period (controls, 6.6%; high dose, 27.3%). Mortality was also slightly higher for high dose males during weeks 52-79 compared to controls, but the increase was not statistically significant (controls, 11.3%; high dose, 15.0%). The percent survival satisfied the requirements for a 2-year chronic study in rats.

TABLE 3: Mortality at various intervals in the 24-month study with DMH

Time period Weeks	Cumulative Mortality (No. rats dead/No. rats alive in the group at the beginning of the time period)			
	Control	100 mg/kg/day	320 mg/kg/day	1000 mg/kg/day
	MALES			
0-52	0/80	1/80	2/80	0/80
52-79	9/80	12/79	11/78	12/80
79-103	39/71	29/67	34/67	40/68
0-103	48/80	42/80	47/80	52/80
% Survival	40	48	41	35
	FEMALES			
0-52	4/80	2/80	1/80	3/80
52-79	5/76	14/78	9/79	21***/77
79-103	27/71	26/64	31/70	20/56
0-103	36/80	42/80	41/80	44/80
% Survival	55	48	48	45

Data extracted from Table 1, pp. 61-68, MRID 44095901.
***$p \leq 0.001$, Significantly different from controls. Fisher exact test conducted by the reviewer.

B. BODY WEIGHT

Group mean body weight gains at various intervals in the study are given in Table 4. There were no significant differences observed in body weights or body weight gains

as a result of DMH treatment in the study. Males tended to lose more weight during the second year of the study than females, but the change was not dose related. Some significant differences in body weight gains were seen sporadically in the treated groups compared to the control groups, but the differences were not consistent and were not dose-related.

TABLE 4: Mean body weight gains at various intervals in the 105-week DMH study.

Time period Weeks	Group mean body weight gain (grams)			
	Control	100 mg/kg/day	320 mg/kg/day	1000 mg/kg/day
	MALES			
0-12	329	331	332	334
12-52	169	164	162	163
52-105	-39	-44	-34	-21
0-105	459	451	460	476
Terminal body weight	626	618	627	643
	FEMALES			
0-12	140	142	142	141
12-52	114	113	108	107
52-105	49	49	35	98
0-105	303	304	285	346
Terminal body weight	440	440	421	482

Data extracted and calculated from Table 5, pp. 164-207, MRID 444095901.

C. FOOD CONSUMPTION AND COMPOUND INTAKE

1. Food consumption

The time-weighted average food consumption levels during various representative weekly time intervals in the study are given in Table 5. The food consumption was slightly increased in treated groups,

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most commonly at the high dose, throughout the study. The increases in the weekly food consumption compared to the control group were often statistically significant ($p \leq 0.05$ to 0.01) during the first year of treatment at the high dose in both sexes, but most commonly in males. No statistically significant differences in weekly food consumption were seen in males after week 86 in treated animals compared to controls, and food consumption in high dose females was slightly increased compared to controls in only 2 weeks (93 and 98 $p \leq 0.05$).

TABLE 5: Mean food consumption at various intervals in the 105-week study with DMH

Time period Weeks	Mean food consumption (g/animal/day)			
	Control	100 mg/kg/day	320 mg/kg/day	1000 mg/kg/day
	MALES			
3 to 4	26	27	27*	28**
4 to 5	27	28*	28*	28**
5 to 6	26	27**	28**	28**
33 to 34	26	27	27	27*
34 to 35	26	26	27*	28**
35 to 36	26	27	27	27*
83 to 84	25	27	27	28**
84 to 85	24	25	25	28**
85 to 86	27	28	26	27
	FEMALES			
3 to 4	19	20	20	20*
4 to 5	19	20	20**	21**
5 to 6	20	20	20	21**
33 to 34	20	21	21	21**
34 to 35	20	20	20	21**
35 to 36	20	20	21	22*
83 to 84	23	22	23	23
84 to 85	22	21	22	22
85 to 86	22	22	22	22

Data extracted from Table 7 pp. 252-293, MRID 44095901.
*p ≤ 0.05, **p ≤ 0.01, significantly different from controls.

2. Compound consumption

The time-weighted average compound consumption for each study group calculated from the body weights and food intake is given in Table 1. The concentration of DMH in the diet was adjusted weekly to achieve

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these doses utilizing body weight and food consumption measurements.

3. Food efficiency

The food efficiency was not calculated for this study.

D. OPHTHALMOSCOPIC EXAMINATION

No treatment-related increases of ocular signs or symptoms were found during the ophthalmoscopic examinations. However, significant decreases in the incidences of bilateral corneal crystals were seen in treated animals compared to the control groups. The changes seen in the incidence of bilateral corneal crystals are shown in Table 6. The incidences of this finding among survivors decreased during the first year and increased again during the second year of treatment in all groups. This change was greater in treated animals especially at the high dose. The decrease at week 51 compared to the controls was statistically significant ($p \leq 0.05$-0.01) in both sexes at the high dose, and was also significant ($p \leq 0.01$) in high dose females at week 103.

TABLE 6: Changes seen in the incidence of bilateral corneal crystals at various times in the 105-week study with DMH

Time period (Weeks)	No. rats with bilateral corneal crystals/No. rats examined/%			
	Control	100 mg/kg/day	320 mg/kg/day	1000 mg/kg/day
	MALES			
Pretest	31/100/31%	14/100/14%	20/100/20%	29/100/29%
Week 51	25/100/25%	18/98/18%	16/98/16%	10**/99/10%
Week 103	13/32/41%	12/39/31%	8/34/24%	11/33/33%
	FEMALES			
Pretest	35/100/35%	25/100/25%	27/100/27%	26/100/26%
Week 51	13/96/14%	13/98/13%	14/99/14%	5*/98/5%
Week 103	20/48/42%	17/38/45%	13/39/33%	5**/36/14%

Data extracted from Table 21, p. 463, Table 22, p 464, and Table 23 p. 465, MRID 44095901.
*p ≤ 0.05, **p ≤ 0.01, significantly different from controls. Fisher exact test conducted by the reviewer.

E. BLOOD WORK

1. Hematology

Although some changes in hematological parameters were identified as statistically significant, they were minor and of little or no biological significance. The red cell count in females was elevated by 8% compared to the control ($p \leq 0.01$) at the high dose in the interim 50-week sample, but the count was not elevated in the main study high dose group at 51 weeks. The mean red cell volume (MCV) was slightly, but consistently, elevated (3-7%, $p \leq 0.01$) at the high dose compared to control values in males at all time intervals in the 105-week oncogenicity subgroup, but there were no significant

differences in the MCV between treated and control groups in the 52-week interim study animals. The increase (6%) of MCV in the high dose males at treatment week 105 was not statistically significant. (Values were taken from Tables 9-14, pp. 336-395, MRID 44095901.)

2. Clinical chemistry

In the one year interim study, serum aspartate aminotransferase in males was elevated ~29% at 320 mg/kg/day ($p \leq 0.05$) and ~34% at 1000 mg/kg/day ($p \leq 0.01$) in treatment week 11 compared to the control group. However, the enzyme activity was decreased by ~25% (NS) and ~28% ($p \leq 0.05$) in the high dose males at treatment weeks 24 and 50, respectively. No additional biologically significant changes were seen in the treated groups compared to the controls in the interim study. In the 105-week study, serum aspartate aminotransferase in high dose males was decreased ~18% ($p \leq 0.05$) at treatment week 12 and was decreased in all treated male groups (~25-30%, $p \leq 0.01$ or 0.05) compared to the control group at week 51. However, aspartate aminotransferase activity in males was not significantly different from the control group at weeks 25, 77, and 105. Aspartate aminotransferase activity was not significantly different from the control activity at any dose level or time point in females. Glucose was slightly (18-29%, $p \leq 0.05$ or 0.01) elevated in high dose males at weeks 12, 25, and 51; and in mid dose males (~21%, $p \leq 0.01$) at 12 weeks. Glucose was slightly elevated in high dose females (17%, $p \leq 0.01$) at treatment week 12 compared to the control group. No additional changes of any biological significance were seen in clinical chemistry parameters in the treated animals compared to the control groups. (Values were taken from Tables 15-16, pp.397-450, MRID 44095901.)

F. URINALYSIS

No dose-related significant changes in urinalysis parameters were seen in the 1-year interim animals. In the 105-week study, urine volume was increased at treatment week 77 in both sexes at the high dose (107% and 136% increase for males and females, respectively, $p \leq 0.05$ or 0.01). The urine specific gravity was reduced in high dose females at week 77 (1.036 and 1.024 for control and high dose, respectively, $p \leq 0.05$). The urine specific gravity was also slightly decreased in high dose males at week 77, but was not statistically significant (1.044 and 1.036 for control and high dose, respectively). (Values were taken from Tables 17-20, pp. 451-462, MRID 44095901.)

G. SACRIFICE AND PATHOLOGY

1. Organ weight

The only statistically significant change in treated animals compared to control animals was a decrease of about 4% in the mean absolute brain weight of high dose interim study males at study week 52. The mean absolute brain weight was also slightly decreased in high dose females at week 52 by about 2%, but the difference was not significant. The slight decrease in mean brain weight, however, contributed to a significant ($p \leq 0.05$) increase in relative kidney (kidney/brain) weight of about 9% in high dose females at week 52 compared to the control relative kidney weight. Absolute brain weight was slightly, but not significantly, decreased in high dose males at the terminal 105-week necropsy. The changes in brain weights and kidney/brain relative weights are given in Table 7.

TABLE 7: Mean absolute and relative (to brain) organ weights after 52 and 105 weeks treatment with DMH

Organ/Treatment period	Mean absolute/relative organ weights (grams)			
	Control	100 mg/kg/day	320 mg/kg/day	1000 mg/kg/day
	MALES			
Brain/52 Week	2.23	2.22	2.19	2.15*
Kidney/52 Week	4.57/204.8	4.64/209.3	4.71/215.1	4.67/216.9
Brain/105 Week	2.27	2.27	2.23	2.21
Kidney/105 Week	5.62/247.2	5.61/246.7	5.58/248.8	5.74/260.0
	FEMALES			
Brain/52 Week	2.01	2.01	1.97	1.97
Kidney/52 Week	2.69/134.3	2.80/139.3	2.70/137.0	2.86/145.9*
Brain/105 Week	2.06	2.04	2.02	2.04
Kidney/105 Week	3.25/157.7	3.47/169.9	3.38/167.4	3.46/170.1

Data extracted from Table 27, pp. 498-501, Table 28, pp. 502-505, Table 31, pp. 514-517, and Table 32, pp. 518-521, MRID 44095901.
*$p \leq 0.05$ Statistically different from controls.

2. Gross pathology

The incidences of gross lesions that may be treatment-related in animals that died or were euthanized moribund in the combined studies are shown in Table 8. The incidences of enlarged pituitary glands in early decedents were significantly ($p \leq 0.05$) increased in high dose males (control, 20%; high dose, 38%) and females (control, 33%; high dose, 61%), and the incidence of reddened pituitary glands was significantly ($p \leq 0.05$) increased in high dose early decedent females (control, 42%; high dose, 56%). The incidence of reddened pituitary glands was also increased in high dose males but the increase was not statistically significant (control, 18%; high dose, 29%). Mammary gland galactoceles were

increased at the high dose in both sexes, but the increase was statistically significant ($p \leq 0.01$) in females (males: control, 10%; high dose 23%; females: control, 31%; high dose, 59%). Increases in gross testicular changes seen in high dose early decedent males were not statistically significant, but can be compared to the findings during the microscopic examinations. A statistically significant increase ($p \leq 0.05$) in the incidence of enlarged iliac lymph nodes was seen in high dose male early decedents. Increases in external surface and subcutaneous abscesses (not statistically significant) in high dose males can be correlated with the enlarge lymph nodes. No statistically significant increases were seen in enlarged lymph nodes or in abscesses in females.

No significant changes in the incidence of gross lesions were seen in the scheduled 52-and 105-week necropsies, and no trends were identified at scheduled necropsies that could be associated with DMH treatment. No gross lesions were seen that correlated with the changes observed in the animals that died prematurely.

TABLE 8: Incidences of gross lesions in early decedents treated with DMH until their death

Organ/Lesion	No. animals with lesion/ No. animals examined/ percent			
	Control	100 mg/kg/day	320 mg/kg/day	1000 mg/kg/day
	MALES			
Pituitary/Reddened	9/50 18%	10/46 22%	10/49 20%	16/56 29%
Pituitary/Enlarged	10/50 20%	11/46 24%	11/49 22%	21*/56 38%
Mammary gland/Galactocele	5/50 10%	6/46 13%	6/49 12%	13/56 23%
Testes/Soft	16/50 32%	12/46 26%	22/49 45%	25/56 45%
Seminal vesicles/Small	8/50 16%	6/46 13%	9/49 18%	17/56 30%
External surface/Abscess	6/50 12%	10/46 22%	4/49 8%	13/56 23%
Subcutaneous/Abscess	1/50 2%	1/46 2%	0/49	6/56 11%
Iliac lymph node/Enlarged	8/50 16%	10/46 22%	8/49 16%	18*/56 32%
	FEMALES			
Pituitary/Reddened	15/36 42%	21/43 49%	15/42 36%	26*/46 56%
Pituitary/Enlarged	12/36 33%	21/43 49%	17/42 40%	28*/46 61%
Mammary gland/Galactocele	11/36 31%	22/43 51%	17/42 40%	27**/46 59%

Data extracted from Table 24, pp. 466-481, MRID 44095901.
*p ≤ 0.05, **p ≤ 0.01, Significantly different from controls.
Fisher exact test performed by reviewer.

3. Microscopic pathology

a) Non-neoplastic - The incidences of lesions found in animals that died or were killed before the end of the DMH treatment period are shown in Table 9. The incidences of selected lesions from rats killed at scheduled times are included in Table 10. The total incidences of animals with selected lesions in the main study (early decedents + 105-week scheduled sacrifice) are shown in Table 11. There were no remarkable effects attributable to DMH treatment at any dose or time period in either early decedents or at scheduled sacrifices. Testicular atrophy was slightly increased at the high dose in early decedent males (control 26%; high dose, 43%, $p = 0.053$) as was testicular fibrinoid vascular degeneration (control, 8%; high dose, 27%, $p \leq 0.01$). This difference was not seen at 52 or 105 weeks. The total incidence of testicular fibrinoid vascular degeneration in the main study high dose males was increased compared to the control group due to the incidence in the early decedents. This lesion was seen in only 2 rats at the high dose and none in the controls or other doses at the 105 week sacrifice. A small increase in nephropathy (NS) and a significant ($p \leq 0.01$) increase in kidney tubular dilatation (control, 65%; high dose, 100%) were seen in males at the high dose compared to the control group in the 52-week interim study. Nephropathy and tubular dilatation were also increased in high dose females, but the differences were not statistically significant or dose-related. The incidences of nephropathy were lower than the controls in both sexes at the high dose at 105 weeks, and the total incidences were slightly, but not significantly, lower in the main study groups. Increased mineralization in the lungs was seen in mid and high dose males at the 52-week scheduled sacrifice (control, 30%; mid dose, 65%, NS; high dose, 68%, $p \leq 0.05$) and in high dose females (control 40%, high dose, 55%, NS). However, the increases in lung mineralization were not seen in either sex at the 105-week sacrifice or the in the total main study animals. Lymphangiectasia of the iliac lymph nodes was significantly increased in both sexes in early decedents and in the total animals in the (main) 105-week study.

TABLE 9: Incidences of non-neoplastic microscopic lesions in animals that died prematurely during DMH treatment

Organ/Lesion	No. Animals with lesion/no. animals examined/%			
	Control	100 mg/kg/day	320 mg/kg/day	1000 mg/kg/day
	MALES			
Testes/Atrophy	13/50/26%	10/46/22%	14/49/29%	24/56/43%
Testes/Fibrinoid vascular degeneration	4/50/8%	3/46/7%	10/49/20%	15**/56/27%
Kidney pelvis/Mineralization	0/50/0%	3/46/7%	4/49/8%	9/56/16%
Iliac lymph node/ Lymphangiectasia	4/50/8%	6/46/13%	8/49/16%	13*/56/23%
–	FEMALES			
Iliac lymph node/ Lymphangiectasia	1/36/3%	0/43/0%	1/42/2%	5/46/11%
Kidney pelvis/Mineralization	13/36/36%	10/43/23%	14/42/33%	15/46/33%

Data extracted from Table 33 pp. 522-589, MRID 44095901.
*p ≤ 0.05, **p ≤ 0.01, Significantly different from controls. Fisher exact test performed by reviewer.

TABLE 10: Incidences of animals with non-neoplastic microscopic lesions at scheduled sacrifices after DMH treatment

Organ/Lesion/Treatment period	No. Animals with lesion/no. animals examined/%			
	Control	100 mg/kg/day	320 mg/kg/day	1000 mg/kg/day
	MALES			
Kidney/Nephropathy/52 Weeks	16/20/80%	14/19/74%	19/20/95%	19/19/100%
Kidney/Tubular dilatation/ 52 Weeks	13/20/65%	16/19/84%	15/20/75%	19**/19/100%
Kidney/Nephropathy/105 Weeks	26/30/87%	30/35/86%	22/31/71%	15*/25/60%
Lungs/Mineralization/52 Weeks	6/20/30%	7/19/37%	13/20/65%	13*/19/68%
	FEMALES			
Kidney/Nephropathy/52 Weeks	6/20/30%	5/20/25%	7/20/35%	7/20/35%
Kidney/Tubular dilatation/ 52 Weeks	5/20/25%	5/20/25%	9/20/45%	7/20/35%
Lungs/Mineralization/52 Weeks	8/20/40%	3/20/15%	8/20/40%	11/20/55%
Kidney/Nephropathy/105 Weeks	14/44/32%	20/37/54%	11/38/29%	6/34/18%

Data extracted from Table 34, pp. 615-642, and Table 35, pp. 647-687,
MRID 44095901.
*p ≤ 0.05, **p ≤ 0.01, Significantly different from controls.
Fisher exact test performed by reviewer.

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TABLE 11: Total Incidences of animals in the oncogenicity study with nonneoplastic microscopic lesions (80 rats/group)

Organ/Lesion/Treatment period	No. Animals with lesion/%			
	Control	100 mg/kg/day	320 mg/kg/day	1000 mg/kg/day
	MALES			
Kidney/Nephropathy	63/79%	59/74%	56/70%	58/73%
Kidney pelvis/Mineralization	2/3%	8*/10%	6/8%	13**/16%
Lungs/Mineralization	1/1%	1/1%	0/0%	1/1%
Iliac lymph node/Lymphangiectasia	10/13%	12/15%	18/23%	21*/26%
Testes/Fibrinoid vascular degeneration	4/5%	3/4%	10/13%	17**/21%
	FEMALE			
Kidney/Nephropathy	23/29%	32/40%	26/33%	15/19%
Kidney pelvis/Mineralization	34/43%	21/26%	31/39%	33/41%
Lungs/Mineralization	0/0%	1/1%	1/1%	2/3%
Iliac lymph node/Lymphangiectasia	1/1%	1/1%	5/6%	7*/9%

Data extracted from Table 33 pp. 522-589, and Table 35, pp. 647-687,
MRID 44095901.
*p ≤ 0.05, **p ≤ 0.01, Significantly different from controls.
Fisher exact test
performed by reviewer.

b) Neoplastic - The number of rats with selected tumors at various times in the 105-week study are given in Table 12. No significant increases in tumor incidences were seen in any treated group compared to the control groups following 105 weeks of treatment with DMH. Pituitary adenomas were the most common lesion seen in these animals at scheduled sacrifices and the most common cause of premature death. Pituitary adenomas were found in 10% of 52-week female controls and in 50% of high dose 52-week females ($p \leq 0.01$); however, the increase was not dose-related. Pituitary adenomas were also increased in high dose 52-week males, but the increase was not

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statistically significant or dose-related (controls, 10%; high dose, 32%). At 105 weeks, there was no difference in the incidence of these tumors between high dose and control animals. Increases were observed in the incidences of

TABLE 12: Number of animals with neoplastic lesions at various times in the 105-week rat study with DMH

Organ/Lesion/Study period	No. Rats with lesion/ No. rats examined/% with lesion			
	Control	100 mg/kg/day	320 mg/kg/day	1000 mg/kg/day
	MALES			
Pituitary, pars distalis/ Adenoma/Predecedents	38/50/76%	32/46/70%	28/48/58%	45/56/80%
Pituitary, pars distalis/ Adenoma/52 Weeks	2/20/10%	6/19/32%	3/20/15%	6/19/32%
Pituitary, pars distalis/ Adenoma/105 Weeks	22/30/73%	21/35/60%	22/31/71%	18/25/72%
Total Incidence with pars distalis adenoma	62/100/62%	59/100/59%	53/99/53%	69/100/69%
External surface/Squamous papilloma/Predecedents	1/50/2%	2/46/4%	1/49/2%	5/56/9%
Total incidence with squamous papilloma	2/80/3%	3/81/4%	3/80/4%	6/81/7%
	FEMALES			
Pituitary, parsdistalis/ Adenoma/Predecedents	30/36/83%	35/42/83%	34/42/81%	42/46/91%
Pituitary, parsdistalis/ Adenoma/52 Weeks	2/20/10%	7/20/35%	4/20/20%	10**/20/50%
Pituitary, parsdistalis/ Adenoma/105 Weeks	42/44/95%	31/37/83%	34/38/89%	31/34/91%
Total incidence with pars distalis adenoma	74/100/74%	73/99/73%	72/100/72%	83/100/83%
Mammary gland/Fibroadenoma/ 105 Weeks	12/44/27%	12/21/57%	9/21/43%	14/34/41%
Total incidence with mammary fibroadenoma	18/80/23%	21/64/33%	16/61/26%	21/80/26%

Data extracted from Table 33A, pp.591-614; Table 34A, pp. 643-646; and Table 35A, pp. 688-701, MRID 44095901.
**p ≤ 0.01, Significantly different from controls. Fisher exact test performed by the reviewer.

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squamous papillomas in early decedent high dose males and in mammary adenofibromas in high dose females at 105 weeks, but these increases were slight, not dose-related, and not statistically significant. No increase in squamous papillomas was seen in treated females at any time point.

III. DISCUSSION

A. DISCUSSION

Dimethylhydantoin (DMH) (97.3%, 97.1% and 93.5% a.i. for lot # 6, # 2412-67-D1, and # 2412-67-D2, respectively) was administered to 80 Crl:CD®BR rats/sex/dose in the diet at dose levels of 0, 100, 320, or 1000 mg/kg/day for 104-105 weeks. The same doses of DMH were given to 20 rats/sex/dose in a 52-week interim study. The concentration of DMH in the diets was adjusted weekly according to body weight and food consumption measurements to achieve the target doses.

Clinical observations included a slightly increased incidence of hypoactivity in high dose animals (males: control, 34%; high dose, 43%, N.S.; females: control, 39%; high dose, 44%, N.S.), and in males, an increased incidence of head tilt (control, 5%; high dose, 18%, $p \leq 0.05$), and impaired equilibrium (control, 6%; high dose, 16%, $p \leq 0.05$). Although these observations are consistent with possible neurological effects, no central nervous system histopathology findings were correlated with the clinical observations. Both sexes had increased incidences of dried yellow matting in the urogenital area at the high dose (males: control, 28%; high dose, 53%, $p \leq 0.001$; females: control, 30%, high dose, 40%, N.S.). The number of times this sign was observed in the daily examinations was increased by 232% in high dose males and 166% in high dose females compared to the control groups.

The overall mortality was slightly, but not significantly, increased in high dose animals (males: control 60%; high dose, 65%; females: control, 45%; high dose, 55%). However, the high dose groups had more deaths early in the study than the control groups. During treatment weeks 52-79, about 11% of male controls died compared to 15% at the high dose. During the same period, about 7% of female controls and 27% ($p \leq 0.001$) of high dose females died. The early deaths, however, were not dose-related.

No significant changes in body weights or weight gains were seen in treated animals compared to controls. The terminal body weights in the high dose groups were slightly higher than in the control groups. This likely reflects a slight increase in food consumption

by the high dose groups. The food efficiency was not calculated.

Changes in hematology were not biologically significant. Glucose was elevated 17%-29% ($p \leq 0.05$-0.01) in high dose males at weeks 12, 25, and 51 and in females at week 12. The periodic elevation of glucose may reflect the increased food consumption seen in high dose animals.

The mean absolute brain weight was slightly (~4%), but significantly ($p \leq 0.05$) decreased compared to the controls in high dose males at 52 weeks. The relative (to brain) kidney weights were slightly increased in high dose males (~6%, N.S.) and high dose females (~9%, $p \leq 0.05$).

No statistically or biologically significant differences were seen in the incidences of gross lesions in the control and treated groups at the scheduled 52- and 105- week sacrifices. However, some differences were seen in early decedents. The incidences of external abscesses and subcutaneous abscesses were increased in high dose males (23% and 11% compared to controls of 12% and 2% for external and subcutaneous abbesses, respectively). The incidence of enlarged iliac lymph nodes was increased in high dose males (control 16%; high dose, 32%, $p \leq 0.05$). Microscopic examination revealed an increased incidence of lymphangiectasia in these lymph nodes in high dose males (control, 8%; high dose, 23%, $p \leq 0.05$). Increases in the number of early decedent high dose males and females with reddened and/or enlarged pituitary glands were seen (reddened pituitary, males: control, 18%; high dose, 29%, N.S.; females: control, 42%; high dose 56%, $p \leq 0.05$; enlarged pituitary males: control, 20%; high dose, 38%, $p \leq 0.05$; females: control, 33%; high dose, 61%, $p \leq 0.05$). Increased incidences of mammary gland galactoceles were also seen in high dose males (control, 10%; high dose, 23%, N.S.) and females (control, 31%; high dose, 59%, $p \leq 0.01$). High dose males also experienced increased incidences of soft testes (control, 32%; high dose, 45%, N.S.) and small seminal vesicles (control, 16%; high dose, 30%, N.S.). Microscopic examination showed increased incidences of testicular atrophy (control, 26%; high dose, 43%, N.S.) and fibrinoid vascular degeneration (control, 8%; high dose 27%, $p \leq 0.01$) in the testes of high dose early decedent males. High dose early decedent males also had an increased incidence of kidney pelvis mineralization (control, 0%; high dose, 16%). The incidences of kidney pelvis mineralization were higher in all female dose groups and controls (23-36%), but the incidences were not dose-related and the differences were not statistically significant.

Microscopic examination at the scheduled 52- and 105-week sacrifices revealed some slight changes in the kidney and lungs of high dose animals compared to the controls. Kidney tubular dilatation at 52 weeks was 100% in high dose males compared to 65% in the control group ($p \leq 0.05$). No difference was seen between the high dose and control groups at 105 Weeks. Lung mineralization was increased in both sexes at the high dose (males: control, 30%; high dose, 68%, $p \leq 0.05$; females: control, 40%; high dose, 55%, N.S.).

No remarkable effects attributable to treatment with DMH occurred in this study, and a definitive target organ or tissue can not be objectively isolated. It is possible that disruptions in pituitary hormones caused secondary effects at the high dose including increased mammary galactoceles in both sexes of early decedents, the high volume and low specific gravity of urine during week 77, increased yellow matting in the urogenital area, increased kidney tubule dilatation in males at 52 weeks, and testicular effects in male early decedents.

A No-Observed-Effect-Level (NOEL) of 320 mg/kg/day was identified. Lowest-Observed-Effect-Levels (LOEL) of 1000 mg/kg/day) for males and females was determined. The LOEL was based on a number of minimal effects including increased mortality earlier in the study, especially in females; reddened and/or enlarged pituitaries in early decedents; slightly decreased brain weight in males; increased mammary galactoceles in both sexes; and testicular fibrinoid vascular degeneration in early decedent males. Most of these effects could be secondary to alterations in the release of pituitary hormones.

There were no significant treatment-related increases in neoplastic lesions in the 105-week study. The incidence of mammary gland fibroadenoma was slightly increased in all treated females compared to the control group, but the increases were not significant or dose-related. Pituitary adenomas (pars distalis) were, by far, the most common neoplastic lesions seen in all animals, and were the most common cause of early deaths in all groups in the study. At 105 weeks, there were no differences in the instances of pituitary adenomas in treated and control groups; however, at 52 weeks, 32% (NS) of high dose males and 50% ($p \leq 0.01$) of high dose females had pituitary adenomas compared to 10% of the male and female control groups. Although the incidences of these tumors were not dose-related, the increased incidences in high dose animals correlate with the increases of reddened and enlarged pituitaries in high dose early decedents. Also, more early decedents at the high dose died in the 6 month period

following 52 weeks of treatment than in the control groups. Slight increases in the incidences of squamous papillomas in high dose male early decedents and of mammary gland fibroadenomas in all treated female groups were not statistically significant or dose-related. At 1000 mg/kg/day, the high dose does meet the requirement of the limit dose for non-toxic substances.

B. STUDY DEFICIENCIES

The lack of statistical calculations made it easy to overlook the near marginal effects of DMH treatment seen in the study. Lactic dehydrogenase activity was not measured in the clinical chemistry portion of the study; however, alkaline phosphatase, aspartate aminotransferase, and alanine aminotransferase were measured, and no gross or microscopic lesions were found to correlate with a change in lactic dehydrogenase activity. The addition of summary tables that include the total incidences of non-neoplastic and neoplastic findings for the main study groups would have been helpful. Historic control data could possibly help show if the marginal increase in pituitary adenomas in high dose rats at 52 weeks was real.

These deficiencies do not detract significantly from the value of the oncogenicity study.

DP BARCODE: D229526 REREG CASE # 305

CASE: 800363 DATA PACKAGE RECORD DATE: 07/08/9
SUBMISSION: S511040 BEAN SHEET Page 1 of 1

* * * CASE/SUBMISSION INFORMATION * * *

CASE TYPE: REREGISTRATION ACTION: 627 CORE DATA
CHEMICALS: 006315 1-Bromo-3-chloro-5,5-dimethylhydantoin 100.00

ID#: 006315-005785
COMPANY: 005785 GREAT LAKES CHEM CORP
PRODUCT MANAGER: 51 KATHLEEN DEPUKAT 703-308-8587 ROOM: CS1 4F6
PM TEAM REVIEWER: PATRICIA LEOPARD 703-308-8065 ROOM: CS1 3RD F
RECEIVED DATE: 09/05/96 DUE OUT DATE: 01/03/97

* * * DATA PACKAGE INFORMATION * * *

DP BARCODE: 229526 EXPEDITE: N DATE SENT: 09/05/96 DATE RET.: / /
CHEMICAL: 006315 1-Bromo-3-chloro-5,5-dimethylhydantoin
DP TYPE: 999 Miscellaneous Data Package
CSF: N LABEL: N

ASSIGNED TO	DATE IN	DATE OUT	ADMIN DUE DATE: 01/03/97
DIV : HED	09/10/96	/ /	NEGOT DATE: / /
BRAN: TB-1	09/12/96	/ /	PROJ DATE: / /
SECT: RS-2	09/25/96	/ /	
REVR : RLOCKE	09/26/96	/ /	
CONTR: OAK RIDGE	09/30/96	/ /	

* * * DATA REVIEW INSTRUCTIONS * * *

GDLN	MRID	Description
83-1(a)	44095901	combined 24-month toxicity
83-2(a)	44095901	study in rats with DMH.

If you have any questions please call me on 308-8065.

Thanks for your help. Patty Leopard.

* * * DATA PACKAGE EVALUATION * * *

* * * ADDITIONAL DATA PACKAGES FOR THIS SUBMISSION * * *

DP BC	BRANCH/SECTION	DATE OUT	DUE BACK	INS	CSF	LABEL