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

SEP 13 1993

SEP 13 1993

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

**MEMORANDUM**

**SUBJECT:** EPA ID 018301; Chlorpropham; CIC DER on an Oncogenicity Study in Mice with Chlorpropham (MRID# 425303-01).

Tox.Chem. No.: 510A.
PC No.: 018301.
DP Barcode No.: D184753.
Submission No.: S429537.
Case No.: 818637.
Action: 627 Generic Data Submission.

From: David G Anderson, PhD
Section 3
Toxicology Branch-1
Health Effects Division (H7509C)

David G Anderson 8/31/93

To: Walter Waldrop/Venus Eagle PM-71
Reregistration Branch
Special Review and
Registration Division (H7508C)

Thru: Karen Hamernik, PhD.
Section 3 Head, Toxicology Branch-1
Health Effects Division (H7509C)

K. Hamernik 8/31/93

KH 9/3/93

**A. CONCLUSIONS:**

Chlorpropham was administered in the feed and adjusted to a constant dose level in mg/kg/day to 50 CD-1 mice per sex per group at 0, 100, 500 or 1000 mg/kg/day for 18-months and 10 mice per sex per group for the 52-week sacrifice.

NOEL: (Systemic) 100 mg/kg/day.

LEL: (Systemic) 500 mg/kg/day for increased hemosiderosis in the spleen and increased hematopoiesis in spleen, liver and bone marrow in males and females. This is consistent with destruction or loss of RBCs. Remarkably dark eyes and bluish extremities were noted and were consistent with possible methemoglobinemia. In addition, at 1000 mg/kg/day the percentage of reticulocytes for males and females was increased. Increases occurred in MCH and MCHC for males and females. Decreased survival occurred in males. Increased spleen and liver weight

were noted in males. This also is consistent with the destruction or loss of RBCs.

The test material was not found to be carcinogenic in this study.

Core classification: Minimum. The study is acceptable under guideline 83-2b for oncogenicity in the mouse.

**B. ACTION REQUESTED:**

Review the reported toxicology in the 18-month oncogenicity in mice with chlorpropham (MRID# 425303-01).

CM 18-month onco study in mice/D184753/425303-01/
A:\CHLORV25.10A\CMONCMOU/DANDERSON/8/31/93*.



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FINAL

# DATA EVALUATION REPORT

Chlorpropham

Oral Oncogenicity Study in Mice

Prepared for:

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July 8, 1993

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Date: 7/16/93

# DATA EVALUATION REPORT

STUDY TYPE: Oral oncogenicity study in mice

TEST MATERIAL: Chlorpropham

TOX CHEM. NUMBER: 510A

PC NUMBER: 018301

MRID NUMBER: 425303-01

SYNONYMS: Isopropyl N-(3-chlorophenyl) carbamate

STUDY NUMBER: 393K-002-050-89

SPONSOR: Chlorpropham Task Force, John Wise and Associates, Ltd., Liberty, MO 64068

TESTING FACILITY: T.P.S., Inc., Mt. Vernon, IN 47621

TITLE OF REPORT: 18-Month Oncogenicity Evaluation of Chlorpropham in the Mouse

AUTHOR: J.A. Botta, Jr.

REPORT ISSUED: October 21, 1992

CONCLUSIONS: Chlorpropham was fed in the diets to CD-1 mice for 18 months at dietary levels to provide a test material intake of about 0, 100, 500, or 1000 mg/kg/day.

NOEL (systemic)=100 mg/kg/day.

LEL (systemic)=500 mg/kg/day based on increased hemosiderosis of the spleen and increased hematopoiesis in the spleen, liver, and bone marrow in both sexes in response to destruction or loss of erythrocytes. Dark eyes and bluish extremities (at limit dose) were also noted. DCH 8/31/93

In addition, at the highest dose tested (1000 mg/kg/day), an increase in percent reticulocytes was seen in males at 12 and 18 months and in females at 12 months; this was accompanied by an increase in MCH and MCHC in both sexes. In addition, survival was significantly lower in males receiving 1000 mg/kg/day than in controls. High-dose males had increased spleen and liver weight. The test material was not found to be [redacted] Carcinogenic in this study. 1

CORE CLASSIFICATION: Core Minimum. The study fulfills the minimum requirements set forth under EPA FIFRA Guideline Series 83-2 for an oncogenicity study in rodents. It does not meet guideline requirements since all kidneys of animals in the mid- and low-dose groups were not examined. This deficiency does not affect the interpretation of the study results.

A. MATERIALS AND METHODS

1. Test Article Description

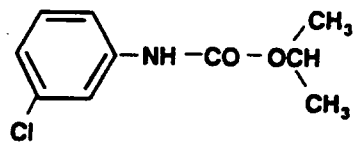
Name: Chlorpropham

Lot number: 14065 L 89

Purity: 96.2% ± 2.0%

Physical property: White crystalline solid

Chemical structure:




Storage: Room temperature in original closed containers

Stability: Not reported (indicated to be the responsibility of Task Force)

2. Diet Preparation

The test material, which was crystalline, was allowed to melt to a liquid at 45-47°C (16 hours). An appropriate amount of test material (corrected to 100% purity) was weighed and dissolved in Mazola corn oil with magnetic stirring. Premixes were prepared by adding the corn oil solution of test compound to about 700-900 g of feed (Purina Certified Rodent Chow Meal #5002) in a Hobart mixer. The beaker that contained the solution was rinsed with corn oil and the contents added to the mixer. The solution and the feed were then mixed for 15 minutes. Diets were prepared weekly for the first 37 weeks and every other week thereafter. The dietary levels in ppm were adjusted as frequently as needed based on food consumption data from the previous week to maintain the desired compound intake on a mg/kg body weight basis. Samples were taken at three levels of the mixer at 11 study intervals for determination of homogeneity and content of test materials. Stability of the test compound in diets over 7 days of storage at ambient temperature was checked at two study intervals.

Results: Most of the analyzed diets were within 5% of the target level in ppm. The test compound was completely stable in diets over a 7-day storage period at ambient temperature. At the four intervals when stability was tested, the ranges analyzed before storage were 99.8-108% of target, and after storage, the analyzed values ranged from 96.9-106% of target. Homogeneity of samples in the dietary mixes was acceptable. The achieved doses as

chlorpropham on a mg/kg basis exceeded the target by >10% at several intervals during the first 6 weeks of the study, but between weeks 13 and 78, achieved levels were closer to the target. Between weeks 53 and 78, 27% of the achieved dosage exceeded ± 10% of the attempted dose.

3. Animals

Species: Mouse

Strain: CD-1

Age: Approximately 4 weeks old at initiation

Weight at initiation: Weights ranged from 21.6 to 31.6 g for males and from 18.9 to 31.1 g for females.

Source: Charles River Laboratories, Inc., Portage, MI

Group assignment: Animals were acclimated to laboratory conditions for 14 days, and healthy, symptom-free animals were assigned to the following groups using computer-generated random number lists.

| Test Group | Doses (mg/kg body wt./day) | Main Study (78 weeks) | | Interim Sacrifice (52 weeks) | |
|---|---|---|---|---|---|
| | | Males | Females | Males | Females |
| 1 Control | 0 | 50 | 50 | 10 | 10 |
| 2 Low-dose (LDT) | 100 | 50 | 50 | 10 | 10 |
| 3 Mid-dose (MDT) | 500 | 50 | 50 | 10 | 10 |
| 4 High-dose (HDT) | 1000 | 50 | 50 | 10 | 10 |

The mice were housed individually in stainless steel animal cages in an isolated temperature- and humidity-controlled animal room (temperature and percent humidity were not stated or reported) with 10-15 air changes/hour and a 12-hour light/dark cycle. Food (Purina Certified Rodent Chow Meal #5002) and water were provided *ad libitum*.

Rationale for dose selection: The dose levels for this study were established by the sponsor based on previous studies.

4. Statistics

Numerical data (body weight, food consumption, hematology parameters, and organ weights) were subjected to analysis of variance followed by Dunnett's test for pairwise comparison of groups or for multiple group comparisons. Survival data were analyzed by Kaplan-Meier life table techniques. The life tables were evaluated with the Cox-Tarone binary regression methods or Gehan-Breslow nonparametic methods. Tumor incidence data were

analyzed by the Cochran-Armitage trend test and by the Fisher-Irwin exact test for pairwise comparison of dosed groups with controls. Survival- adjusted tumor analysis was performed as necessary when there were group differences in intercurrent mortality. Incidental tumors were analyzed by logistic regression of tumor prevalence.

5. Quality Assurance

A signed quality assurance statement, dated October 21, 1992, was provided. A GLP certification statement and a flagging statement were present.

B. METHODS AND RESULTS

1. Clinical Observations

All animals were observed twice daily for clinical signs of toxicity, physical appearance, and behavioral changes. Animals that were dead or moribund were removed and the date and disposition recorded. Animals were examined in detail individually once a week for the first 13 weeks and biweekly thereafter, and they were palpated for masses.

Results: Table 1 presents data for mortality and percent survival at selected intervals during the study. At termination (78 weeks), survival ranged from 58% to 88% in male groups and from 62% to 78% in female groups. Mortality in high-dose males at week 78 was significantly higher than in controls. The major cause of death was reported to be amyloidosis.

The most frequent clinical observations were a bluish tint to the skin of the extremities and dark eyes. These changes occurred in high-dose males and females as early as 4 weeks. In the mid-dose groups, a bluish tint to the skin was observed in 11 males and 4 females by week 34. By study termination, dark eyes were observed in all high-dose males and females, in 58/60 (96.7%) mid-dose males, and in 19/60 (31.7%) mid-dose females, and bluish skin was observed in all high-dose animals, 13% of the mid-dose females, and 40% of the mid-dose males. No increases in the incidence of masses was observed in dosed groups. Other clinical observations were considered incidental.

2. Body Weights/Food and Water Consumption/Test Material Intake

Body weights and food consumption (per cage) were recorded weekly for the first 13 weeks and biweekly thereafter. Cumulative weight gains and compound intake were calculated at the same intervals.

Results: No consistent or treatment-related changes in either mean body weight or mean body weight change related to dosing were observed. No treatment-related effects on mean food consumption values were observed (data not shown). The mean compound intake over the course of the study was $108.9 \pm 9.2$ mg/kg/day, $523.0 \pm 42.0$ mg/kg/day, and $1050.6 \pm 82.8$ mg/kg/day on a body weight basis in males in the low-, mid-, and high-dose groups,

Table 1. Mortality and (Percent Survival) in Mice Fed Chlorpropham for 18 Months

| Dosage Level (mg/kg/day) | Mortality and (Percent Survival) at Week | | | | |
|---|---|---|---|---|---|
| | 13 | 26 | 52 | 65[a] | 78[a] |
| | | | Males | | |
| 0 | 0 (100) | 1 (98) | 2 (97) | 3 (94) | 7 (86) |
| 100 | 0 (100) | 0 (100) | 2 (97) | 3 (94) | 10 (80) |
| 500 | 0 (100) | 0 (100) | 1 (98) | 1 (98) | 6 (88) |
| 1000 | 4 (93) | 4 (93) | 7 (88) | 11 (79) | 22 (58) |
| | | | Females | | |
| 0 | 0 (100) | 0 (100) | 2 (97) | 5 (90) | 8 (84) |
| 100 | 1 (98) | 1 (98) | 8 (87) | 8 (85) | 17 (68) |
| 500 | 1 (98) | 2 (97) | 4 (93) | 7 (86) | 17 (67) |
| 1000 | 0 (100) | 0 (100) | 1 (98) | 3 (94) | 11 (78) |

Source: Study No. 393K-002-050-89, Table 4.

[a]Percent survival at weeks 65 and 78 is based on denominators of 50, 51, 50, and 52 for males and 50, 53, 51, and 50 for females at increasing doses since at the interim sacrifice 10, 9, 10, and 8 males at doses of 0, 100, 500, and 1000 mg/kg/day and 10, 7, 9, and 10 females at the same doses were sacrificed, rather than 10/sex/group as specified by the protocol. For weeks 13, 26, and 52, N is 60/sex/group.



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respectively, and 108.1±11.5 g, 518.6±46.9 g, and 1060.4±85.8 g in females in the same groups.

3. Ophthalmoscopic Examination

Ophthalmoscopic examinations were conducted on all animals prior to initiation, on 10 mice/sex/group prior to the interim sacrifice, and on all survivors prior to the terminal sacrifice.

Results: Pigment in the cornea was observed in 3/10 mid-dose males and 3/10 high-dose males at week 52; in females, pigment in the cornea was observed at 52 weeks in 1/10 controls and 1/10, 2/10, and 4/10 animals in the low-, mid-, and high-dose groups, respectively. However, prior to the terminal sacrifice, the incidences of mice with pigment deposits in the cornea were 19%, 20%, 12%, and 14% in males and 12.5%, 8%, 0%, and 5% in females at doses of 0, 100, 500, or 1000 mg/kg/day, respectively, suggesting that increases at 12 months were not treatment related.

4. Clinical Pathology

Blood was drawn from 10 mice/sex/group prior to the interim and terminal sacrifices, and the hematology parameters checked (X) below were examined.

(a) Hematology

| | |
|---|---|
| X Hematocrit (HCT) | X Leukocyte differential count |
| X Hemoglobin (HGB)* | X Mean corpuscular HGB (MCH) |
| X Leukocyte count (WBC)* | X Mean corpuscular HGB concentration (MCHC) |
| X Erythrocyte count (RBC)* | X Mean corpuscular volume (MCV) |
| X Platelet count* | Coagulation: thromboplastin time (PT) |
| X Reticulocyte count (RETIC) | |
| Red cell morphology | |

*Recommended by Subdivision F (November 1984) Guidelines

Results: Selected hematology parameters at 12 and 18 months are summarized in Table 2. Reticulocyte counts were significantly increased ($p=0.01$) in mid- and high-dose males at 12 months and in high-dose males at 18 months. Reticulocyte counts were also increased in high-dose females at 12 months but not at 18 months. At 12 months, mean corpuscular hemoglobin (MCH) was increased compared to controls in high-dose males and high-dose females and MCHC was increased in high-dose males. At 18 months, MCH was significantly increased in high-dose males and MCHC was significantly increased in high-dose females. No changes in any other hematology parameters were considered related to treatment. The increase in reticulocytes would indicate a compensatory mechanism to a destruction of erythrocytes. The increases in MCH and MCHC would follow maturation of reticulocytes to mature erythrocytes.

Table 2. Mean Values (±SD) for Selected Hematology Parameters in Mice Fed Chlorpropham for 18 Months.

| Parameter/ Interval | Dosage Level (mg/kg/day) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Males | | | | Females | | | |
| | 0 | 100 | 500 | 1000 | 0 | 100 | 500 | 1000 |
| MCH (pg) | | | | | | | | |
| 12 mos. | 16.1±0.9 | 15.9±0.9 (98.8)[a] | 16.4±0.6 (101.9) | 17.0±0.4* (105.6) | 16.3±1.0 | 16.6±0.7 (101.8) | 16.5±0.9 (101.2) | 17.3±0.9* (106.1) |
| 18 mos. | 16.5±0.9 | 16.5±0.7 (100.0) | 17.2±0.4 (104.2) | 18.0±1.4** (109.1) | 17.3±1.5 | 17.4±1.3 (100.6) | 16.9±0.5 (97.7) | 17.7±0.9 (102.3) |
| MCHC (g/dL) | | | | | | | | |
| 12 mos. | 36.1±1.0 | 36.4±0.6 (100.8) | 36.5±0.8 (101.1) | 37.7±1.1** (104.4) | 36.6±0.6 | 36.2±0.7 (98.9) | 36.5±1.0 (99.7) | 37.0±0.8 (101.1) |
| 18 mos. | 36.9±0.9 | 36.7±1.0 (99.5) | 37.6±0.7 (101.9) | 38.0±1.7 (103.0) | 36.0±1.6 | 37.0±1.1 (102.8) | 37.1±0.7 (103.1) | 37.8±0.8** (105.0) |
| Reticulocyte (%) | | | | | | | | |
| 12 mos. | 1.8±0.8 | 2.1±0.7 (116.7) | 3.1±1.1** (172.2) | 3.7±0.8** (205.6) | 1.4±0.5 | 1.9±0.7 (135.7) | 2.5±0.7 (178.6) | 4.2±4.2* (300.0) |
| 18 mos. | 1.1±0.7 | 1.3±0.8 (118.2) | 1.8±1.0 (163.6) | 4.2±4.3** (381.8) | 3.3±3.8 | 2.5±1.4 (75.8) | 2.6±1.2 (78.8) | 3.1±1.3 (93.9) |

Source: Study No. 393K-002-050-89, Table 10.

[a]The values given in parentheses represent percent of control values for each given parameter.

*Significantly different from control value, p=0.05.
**Significantly different from control value, p=0.01.

(b) Blood (Clinical) Chemistry

Clinical chemistry parameters were not examined.

(c) Urinalysis

Urinary parameters were not examined.

5. Sacrifice and Pathology

All animals that died or were sacrificed moribund and all animals sacrificed at 53 weeks or at study termination received a complete necropsy. The tissues checked (X) below were collected and preserved in formalin, and the double-checked (XX) organs were weighed at the interim and terminal sacrifices. All tissues from the animals that died or were sacrificed moribund, as well as target organs and any gross abnormalities from mid- and low-dose groups, were also examined microscopically.

| | | | | | |
|---|---|---|---|---|---|
| Digestive System | | Cardiovascular/Hematologic | | Neurologic | |
| | Tongue | X | Aorta* | XX | Brain* (three levels) |
| X | Salivary glands* | XX | Heart* | X | Peripheral nerve* (sciatic nerve) |
| X | Esophagus* | X | Bone marrow* | X | Spinal cord* (three levels) |
| X | Stomach* | X | Lymph nodes* | XX | Pituitary* |
| X | Duodenum* | XX | Spleen* | X | Eyes* (Optic nerve) |
| X | Jejunum* | X | Thymus* | | |
| X | Ileum* | | | Glandular | |
| X | Cecum* | Urogenital | | | |
| X | Colon* | | | XX | Adrenals* |
| X | Rectum* | XX | Kidneys* | | Lacrimal gland |
| XX | Liver* | X | Urinary bladder* | X | Mammary gland |
| X | Gallbladder* | XX | Testes* | XX | Thyroids* with parathyroids* |
| X | Pancreas* | XX | Epididymides | X | Harderian glands |
| Respiratory | | X | Prostate* | | |
| | | X | Seminal vesicle | | |
| X | Trachea* | XX | Ovaries* | | |
| X | Lungs* | X | Uterus* | | |

Other

X Bone (sternum and femur)*
X Skeletal muscle*
X Skin*
X All gross lesions and masses*

*Recommended by Subdivision F (November 1984) Guidelines

(a) Organ Weights

Table 3 presents data for liver and spleen weights. The mean liver weights and liver weights as percent of body weight were significantly (p=0.05 or p=0.01) increased (24%) in high-dose males at 12 months. The absolute liver weights in high-dose males were also significantly (p=0.05) increased at 18 months (11% when compared to controls); the relative liver weights (as percent of body weight) in high-dose males were nonsignificantly increased at 18 months (8%). No significant increases in absolute or relative liver weights were seen in dosed females. Spleen weights (absolute and relative) were significantly increased (85-87%) in high-dose males at termination (p=0.01); slight increases in spleen weights (absolute and relative) were seen for high-dose males and females at 12 months, 66% and 50%, respectively, when compared to controls. The spleen weights increases in males appear to be treatment related in the high dose. No effects of dosing were apparent on the weights of other organs.

(36 and 39 % resp (12 months)

(b) Macroscopic Pathology

No distinct or consistent treatment-related organ or tissue changes were seen at the interim or terminal necropsies or for animals found dead or sacrificed moribund.

(c) Microscopic Pathology

Nonneoplastic lesions: Table 4 summarizes data for findings in the spleen, liver, and bone marrow of mice. An increase in hematopoiesis (primarily erythropoiesis) was seen at all three sites in groups receiving or 1000 mg/kg/day. Increased cellularity of the bone marrow at 500 and 1000 mg/kg/day was also seen in both sexes at the mid-dose level.

At the 12-month sacrifice, no mice had moderate or severe hematopoiesis in the spleen. Hematopoiesis, graded slight-to-moderate was present in all high-dose males and females (10/sex); 13/20 mid-dose mice and 2/20 low-dose mice had slight hematopoiesis, whereas the remaining mice had minimal hematopoiesis. Minimal hematopoiesis is a normal finding and slight hematopoiesis is of equivocal toxicologic importance. In the main groups, there was an increase in marked or moderate hematopoiesis in the spleen (weeks 53-78). Slight hematopoiesis was present in 12%, 14%, 36% and 60% of the males at 0, 100, 500, or 1000 mg/kg/day and moderate or marked hematopoiesis was seen in 2%, 0%, 8%, and 18% of males in the same dose groups; the remaining mice had minimal hematopoiesis (normal finding). For females at the same doses, 30%, 30%, 52%, and 70% had slight hematopoiesis in the spleen and 8%, 0%, 6%, and 14% of the females receiving 0, 100, 500, or 1000 mg/kg/day had marked or moderate hematopoiesis in the spleen. Hemosiderosis (minimal/slight) in the spleen was increased in incidence in both sexes at 500 and 1000 ppm (Table 4).



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Table 3. Mean Organ Weights (±SD) and Relative Organ Weights (as % of Body Weight) for Liver and Spleen of Mice Administered Chlorpropham in the Diet

| Organ/ Interval | Dosage Level (mg/kg/day) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Males | | | | Females | | | |
| | 0 | 100 | 500 | 1000 | 0 | 100 | 500 | 1000 |
| Liver | | | | | | | | |
| Interim sacrifice | | | | | | | | |
| (g) | 1.44±0.17 | 1.49±0.15 | 1.59±0.22 | 1.79±0.37* | 1.25±0.22 | 1.46±0.08 | 1.46±0.21 | 1.43±0.34 |
| (%) | 4.46±0.42 | 4.42±0.43 | 4.84±0.75 | 5.54±0.78** | 4.40±0.53 | 4.74±0.28 | 4.62±0.55 | 4.96±0.68 |
| Terminal sacrifice | | | | | | | | |
| (g) | 1.48±0.30 | 1.45±0.21 | 1.56±0.27 | 1.65±0.20* | 1.61±0.05 | 1.56±0.30 | 1.53±0.22 | 1.62±0.32 |
| (%) | 4.83±1.05 | 4.63±0.61 | 5.00±0.74 | 5.22±0.67 | 5.14±1.11 | 5.13±0.77 | 5.14±0.63 | 5.37±1.05 |
| Spleen | | | | | | | | |
| Interim sacrifice | | | | | | | | |
| (g) | 0.11±0.05 | 0.09±0.04 | 0.11±0.04 | 0.15±0.09 | 0.10±0.02 | 0.11±0.06 | 0.11±0.03 | 0.15±0.08 |
| (%) | 0.33±0.15 | 0.29±0.11 | 0.32±0.12 | 0.46±0.23 | 0.34±0.07 | 0.36±0.18 | 0.36±0.11 | 0.51±0.20* |
| Terminal sacrifice | | | | | | | | |
| (g) | 0.08±0.03 | 0.09±0.04 | 0.10±0.04 | 0.15±0.12** | 0.17±0.17 | 0.14±0.08 | 0.12±0.04 | 0.15±0.05 |
| (%) | 0.26±0.10 | 0.30±0.14 | 0.33±0.12 | 0.49±0.40** | 0.52±0.48 | 0.45±0.25 | 0.40±0.12 | 0.51±0.17 |

Source: Study No. 393K-002-050-89, Table 11.

*Significantly different from control value, p=0.05.
**Significantly different from control value, p=0.01.

Table 4. Nonneoplastic Findings in Spleen, Liver, and Bone Marrow of Mice Fed Chlorpropham[a]

| Organ/ Finding/ Severity | Dosage Level (mg/kg/day) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Males | | | | Females | | | |
| | 0 | 100 | 500 | 1000 | 0 | 100 | 500 | 1000 |
| Spleen | (59)[b] | (59) | (60) | (59) | (60)[d] | (60) | (60) | (60) |
| **Hematopoiesis**[c] | | | | | | | | |
| Slight | 8 | 7 | 26 | 39 | 15 | 17 | 31 | 42 |
| Moderate | 2 | 0 | 4 | 10 | 4 | 0 | 6 | 6 |
| Marked | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 4 |
| **Hemosiderosis** | | | | | | | | |
| Minimal/slight | 2 | 1 | 32 | 43 | 11 | 15 | 31 | 57 |
| Liver | (59) | (60) | (60) | (59) | (60) | (60) | (60) | (60) |
| **Hematopoiesis** | | | | | | | | |
| Minimal/slight | 0 | 0 | 1 | 9 | 0 | 1 | 2 | 7 |
| Bone Marrow[c] | (59) | (60) | (60) | (60) | (60) | (60) | (60) | (60) |
| **Cellularity** | | | | | | | | |
| Marked | 5 | 2 | 21 | 53 | 7 | 4 | 20 | 46 |

Source: Study No. 393K-002-050-89, Tables 16A and 16B and Appendix X, pp. 850-873 (Interim Sacrifice) and pp.873-1034 (Main Study).

[a]Includes animals at interim and terminal sacrifice and those found dead or sacrificed moribund.
[b]The number of animals in each groups with the specific organ examined histologically
[c]All animals had some grade of hematopoiesis in the spleen and those not having marked cellularity in bone marrow had moderate cellularity. (Minimum, slight, moderate, marked)
[d]One control female that died in week 1 was replaced. Data for this animal were deleted in this tabulation.

At 12 months, minimum hematopoiesis of the liver was seen in 3/10 high-dose males and 5/10 high-dose females.

At the interim sacrifice, 19/20 high-dose animals and 8/20 mid-dose animals had marked cellularity of the bone marrow with increased erythropoiesis. At the terminal sacrifice (including deaths from 53 weeks), 30% and 40% of mid- and high-dose males, respectively, and 68% and 72% of females in the same doses had marked cellularity. All mice had some degree of bone marrow cellularity.

The increased incidence and severity of hemosiderosis and hematopoiesis correlate with slight increases in the weight of the liver and spleen at the interim sacrifice and with the reticulocytosis and increased MCH and MCHC levels in the high-dose groups at both 12 and 18 months.

The other nonneoplastic findings recorded were considered to be incidental and not related to dosing with the possible exception of amyloidosis. Amyloidosis at multiple sites (primarily liver, kidneys, adrenal, thyroid, and the gastrointestinal tract) was observed in 16 control males (28.1%) and 19 high-dose males (32.2%) and in 12 control females (20.0%) and 16 high-dose females (26.7%). At the 12-month sacrifice, amyloidosis was infrequent, graded minimal, and considered incidental. Amyloidosis is a common spontaneous finding in older mice. It is most prevalent in the ileum, kidney, and liver but is also found in several other organs. In the high-dose males, it was reported by the study author to be a contributory cause of death or moribundity in 11/20 mice. In high-dose females, amyloidosis was reported to be the major cause of death (9/11 mice versus 2/11 controls).

Neoplastic lesions: No increases in the incidences of neoplasms at any tissue/organ site were observed in either sex when dosed groups were compared with controls. The incidence of all neoplasms was within the normal 18-month range observed for Charles River CD-1 (ICR) mice (Lang, P., Charles River Laboratories, 1991).

C. REVIEWERS' DISCUSSION AND INTERPRETATION OF RESULTS

The study was adequately conducted, and summary tabulations of data were supported by individual animal data. Tabular correlations of masses observed in life and those seen on gross examinations were provided, and the time of first observation was also given so that time to appearance of tumor could be easily determined. Gross and histological correlation data were also tabulated and showed an excellent correlation of masses and confirmed neoplasms. One minor deficiency was noted -- the kidneys were not examined at terminal sacrifice for animals in the mid- and low-dose groups. These data could have been helpful to determine if there was a dose-related trend for exacerbation of amyloidosis by administration of chlorpropham.

No effects of toxicological importance on body weights or body weight gains were observed to support the adequacy of dosing (establish an MTD), however the high dose was at the limit test of 1000 mg/kg/day.

Previous subchronic and chronic studies in mice may have established an MTD; however, none of these studies were available for review. A LOEL and NOEL based on systemic toxicity can be established, however, based on an increase in the incidence of hemosiderosis in the spleen and an increase in hematopoiesis in the spleen and bone marrow at the mid and high doses in both sexes. Although decreases in RBC and HGB were not seen in the study, reticulocyte counts increased in mid-dose males and in mid- and high-dose females, and hemosiderosis in the spleen was increased in the mid- and high-dose groups. It is clear that a destruction or loss of erythrocytes occurred and this was compensated for by an increase in reticulocytes and increased hematopoiesis.

No increase in the incidence of tumors was seen under the conditions of the study. A NOEL of 100 mg/kg/day and a LOEL of 500 mg/kg/day can be established based on increased hemosiderosis in the spleen and increased hematopoiesis in the spleen and bone marrow.