# DATA EVALUATION RECORD

MESOTRIONE (ZA 1296) 11/3/2000

Study Type: §83-1(b), Chronic Oral Toxicity Study in Mice

Work Assignment No. 2-1-52V (MRID 44505026)

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

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

Primary Reviewer
Joan L. Harlin, M.S.

Signature: Joan L Harlin
Date: 3/25/00

Secondary Reviewer
Kelley Van Vreede, M.S.

Signature: Kelley Van Vreede
Date: 3/30/00

Program Manager
Mary L. Menetrez, Ph.D.

Signature: Mary L Menetrez
Date: 3/30/00

Quality Assurance
Steve Brecher, Ph.D.

Signature: Steven Brecher
Date: 3/30/00

Disclaimer

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

1

253

EPA Reviewer: David Nixon, D.V.M.
Registration Action Branch 1/HED (7509C)

David Nixon 10/31/2000

Work Assignment Manager: Marion Copley, D.V.M., D.A.B.T.
Registration Action Branch 1/HED (7509C)

Mcopley 11/3/00

DATA EVALUATION RECORD

STUDY TYPE: One-year chronic toxicity [feeding] - mice
OPPTS Number: 870.4100 OPP Guideline Number: §83-1b

DP BARCODE: D259369 SUBMISSION CODE: S541375
P.C. CODE: 122990 TOX. CHEM. NO.: None

TEST MATERIAL (PURITY): Mesotrione (96.8% a.i.)

SYNONYMS: ZA 1296; 2-[4-(Methylsulphonyl)-2-nitrobenzoyl]-1,3-cyclohexanedione; 2-(4-mesyl-2-nitrobezoyl)-cyclohexane-1,3-dione

CITATION: Pinto, P.J. (1997) ZA 1296: One Year Dietary Toxicity Study in Mice. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK. Laboratory Project Report No. CTL/P/5682. Laboratory Project Study No. PM1062. November 12, 1997. MRID 44505026. Unpublished.

SPONSOR: Zeneca Ag Products, Wilmington, DE

EXECUTIVE SUMMARY: In this chronic oral toxicity study (MRID 44505026), mesotrione (ZA 1296; Batch No. P17; 96.8% a.i.) was administered continuously in the diet to 60 C57BL/10J$_f$CD-1 mice/sex/dose at nominal dose levels of 10, 50, 350, or 7000 ppm (equivalent to 1.5, 7.8, 56.2, or 1114.0 mg/kg/day in males and 2.1, 10.3, 72.4, and 1494.5 mg/kg/day in females) for 3, 6, or 12 months. Two control groups of 60 C57BL/10J$_f$CD-1 mice/sex (120/sex) were fed untreated diet for 3, 6, or 12 months. There were 20 treated mice/sex/group per interval.

Mortality, clinical observations, body weights, food consumption, ophthalmoscopic observations, hematological parameters, clinical chemistry parameters, urinalysis parameters, organ weights, and gross and microscopic pathological findings were unaffected by the test substance.

Body weight gain in the 7000-ppm males was reduced overall by 17%. Reductions in body weight gain appeared to occur throughout the study. In conjunction with a 23% decrease in food utilization during the first 4 weeks of the study, an overall decrease in food utilization of 9%

from weeks 1-13, and no decrease in food consumption throughout the study, the evidence suggests that there was a toxicologically significant effect in the 7000-ppm males.

Under the conditions of this study, there was no evidence of carcinogenic potential.

**The LOAEL for this study is 7000 ppm (equivalent to 1114.0 mg/kg/day in males and 1494.5 mg/kg/day in females) based on decreases in body weight gain and food utilization in males. The NOAEL is 350 ppm (equivalent to 56.2 mg/kg/day in males and 72.4 mg/kg/day in females).**

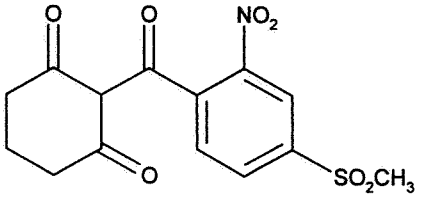
The submitted study is classified as **acceptable/guideline [§83-1(b)]** and satisfies the guideline requirements for a chronic oral toxicity study in rodents.

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

## I. MATERIALS AND METHODS

### A. MATERIALS

1. Test Material: Mesotrione (ZA 1296)
   Description: Light beige solid
   Lot/Batch #: P17
   Purity: 96.8% a.i.
   Stability of compound: The test substance was stable for over 2.5 years when stored at ambient temperature in the dark.
   CAS #: 104206-82-8
   Structure:




2. Vehicle: Diet

3. Test animals: Species: Mouse
   Strain: C57BL/10J$_f$CD-1
   Age and weight at study initiation: Approximately 5-6 weeks old; 17.1-25.4 g (males), 13.6 g- 20.4 g (females)
   Source: Barriered Animal Breeding Unit, Zeneca Pharmaceuticals, Alderley Park
   Housing: Five mice per cage by sex in multiple mice racks.
   Diet: CT1 Diet (Special Diet Services Ltd., Stepfield, Witham, Essex, UK), ad libitum
   Water: Tap water, ad libitum

Environmental conditions:
Temperature: 16-30°C
Humidity: 39-91%
Air changes: At least 15/hour
Photoperiod: 12 hours light:12 hours dark
Acclimation period: Approximately 1 week

B. STUDY DESIGN:

1. In life dates - start: 08/14/96 end: 08/19/97

2. Animal assignment - Animals were randomly assigned (stratified by body weight) to treatment groups as indicated in Table 1.

Table 1. Study design

| Test Group[a] | Conc. in Diet (ppm) | Achieved Dose[b] (mg/kg/day) [M/F] | Males | Females |
|---|---|---|---|---|
| Control | 0 | 0/0 | 60 | 60 |
| Control | 0 | 0/0 | 60 | 60 |
| Low | 10 | 1.5/2.1 | 60 | 60 |
| Low-mid | 50 | 7.8/10.3 | 60 | 60 |
| Mid | 350 | 56.2/72.4 | 60 | 60 |
| High | 7000 | 1114.0/1494.5 | 60 | 60 |

a Twenty mice/sex/group were sacrificed on schedule at 3, 6, or 12 months.
b Achieved dosages (mg/kg/day) for one year of dosing were obtained from the study report Appendix F, pages 157 and 158.

3. Dose selection rationale - Dose selection for the current study was based on the results of shorter term feeding studies conducted in this laboratory (no further information provided). The highest dose level of 7000 ppm selected for the current study is considered to be the limit dose for this strain of mouse.

4. Diet preparation and analysis - Diets were prepared by mixing the appropriate amounts of test substance with the feed and stored in plastic bins at -20°C. The diets were thawed for approximately one hour prior to use. Duplicate samples (top, middle, bottom) from the 10 ppm and 7000 ppm diet formulations prepared 9 days prior to study initiation and during study week 2 were analyzed for homogeneity (analysis dates not reported). The stability of ZA 1296 in the diet was tested during previous studies (PR1001 and PM0983; not further referenced). In those studies, samples of 1 ppm and 7000 ppm diet



4
2586

formulations were analyzed for stability following storage at room temperature for up to 9 and 16 days, respectively, or at -20°C for up to 57 and 40 days, respectively. Concentration was analyzed in all diets prepared for the current study 9 days prior to study initiation and during study week 10.

Concentration analysis (range as mean % of nominal): 99.0-111%

Homogeneity (range as mean % of nominal): 93.4-109%

Stability (% of day 0):
Room temperature: 84.8-107.4%
-20°C: 87.5-114.6%

The analytical data indicate that the mixing procedure was adequate and that the variation between nominal and actual dosage to the study animals was acceptable.

4. Statistics - Body weight data were analyzed for each sex by analysis of covariance. Food consumption and food utilization data were analyzed by analysis of variance (ANOVA). Male and female hematology, blood chemistry, and urinalysis data were analyzed together by ANOVA. Organ weights were analyzed by ANOVA and analysis of covariance. All analysis of variance and analysis of covariance were followed by a Student's t-test, as necessary. Week 53 plasma GGT and plasma chloride were evaluated by ANOVA for males only.

C. METHODS:

1. Observations - All animals were observed daily for changes in clinical condition or behavior. Detailed clinical observations were performed weekly.

2. Body weight - All animals were weighed at study initiation, weekly during weeks 1-14 and 17, then every 4 weeks, and prior to study termination.

3. Food consumption and compound intake - Food consumption was measured for each cage of mice at weekly intervals during weeks 1-13, week 16, and then every 4 weeks throughout the treatment period. A weekly mean (g/animal/day) was calculated for each cage. Food utilization was calculated as body weight gained per cage/100 g food consumed for weeks 1-13 only. Mean compound intake was calculated as mg/kg/day using nominal test concentration, food consumption, and body weight data.

4. Ophthalmoscopic examination - Ophthalmoscopic examinations were performed on all surviving control and high-dose animals the week before termination at the 3 and 12 month time points. The eyes of all surviving females in the mid, low-mid, and low dose groups were examined at 12 months. The ophthalmological data for mice sacrificed at 3 months were reported in MRID 414505022.

5. Blood - Blood, obtained by cardiac puncture, was collected from 10 animals/sex/group at weeks 14, 27, and 53. It was not reported whether the animals were fasted prior to blood collection. The checked (X) hematology and clinical blood chemistry parameters 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 corpusc. HGB conc.(MCHC) |
| X | Erythrocyte count (RBC) | X | Mean corpusc. volume (MCV) |
| X | Platelet count | | Reticulocyte count |
| | Blood clotting measurements | | |
| | (Thromboplastin time) | | |
| | (Activated partial thromboplastin time) | | |
| | (Clotting time) | | |
| | (Prothrombin time) | | |

b. Clinical Chemistry

| | ELECTROLYTES | | OTHER |
|---|---|---|---|
| X | Calcium | X | Albumin |
| X | Chloride | X | Blood creatinine |
| | Magnesium | X | Blood urea nitrogen |
| X | Phosphorus | | Total Cholesterol |
| X | Potassium | | Globulin |
| X | Sodium | X | Glucose |
| | | X | Total bilirubin |
| | ENZYMES | X | Total serum protein (TP) |
| X | Alkaline phosphatase (ALK) | | |
| | Cholinesterase (ChE) | | |
| X | Creatine kinase | | |
| | Lactic acid dehydrogenase (LDH) | | |
| X | Serum alanine aminotransferase (also SGPT) | | |
| X | Serum aspartate aminotransferase (also SGOT) | | |
| X | Gamma glutamyl transferase (GGT) | | |
| | Glutamate dehydrogenase | | |

6. Urinalysis - Urine was collected from each cage of surviving mice over a 10-18 hour period during weeks 13, 26, and 52. All animals had access to water; females also had access to food. The checked (X) parameters were examined.

| | | | |
|---|---|---|---|
| X | Appearance (including color) | X | Glucose[a] |
| X | Volume | X | Ketones[a] |
| X | Specific Gravity | X | Bilirubin[a] |
| X | pH | X | Blood[a] |
| X | Sediment (microscopic) | | Nitrite |
| X | Protein[a] | | Urobilinogen |

a Assessed semi-quantitatively only.

7. Sacrifice and Pathology - All mice were killed by exsanguination upon their scheduled termination. Ten mice/sex/group were subjected to a gross pathological examination at weeks 14 and 53. The following CHECKED (X) tissues were collected and preserved in appropriate fixative; the (XX) organs were weighed. All (X) tissues except the oral and nasopharyngeal cavities from the control and high-dose animals and all animals killed prematurely were examined histologically. Additionally, the lungs, liver, gall bladder, and kidneys were examined microscopically in all animals. The adrenal glands were examined microscopically in all males. Week 14 organ weight and pathology data were reported in MRID 44505022.

| | DIGESTIVE SYSTEM | | CARDIOVASC./HEMAT. | | NEUROLOGIC |
|---|---|---|---|---|---|
| | Tongue | X | Aorta | XX | Brain (cerebrum, cerebellum, |
| X | Salivary glands | X | Heart | | and brainstem) |
| X | Esophagus | X | Bone marrow | X | Periph. nerve |
| X | Stomach | X | Lymph nodes | X | Spinal cord |
| X | Duodenum | X | Spleen | | (cervical, thoracic, lumbar) |
| X | Jejunum | X | Thymus | X | Pituitary gland |
| X | Ileum | | _____ | X | Eyes |
| X | Cecum | | UROGENITAL | | _____ |
| X | Colon | XX | Kidneys | | GLANDULAR |
| X | Rectum | X | Urinary bladder | XX | Adrenal gland |
| XX | Liver | XX | Testes | X | Harderian gland |
| X | Gall bladder | X | Epididymides | | Lacrimal gland |
| X | Pancreas | X | Prostate | X | Mammary gland |
| X | Oral cavity* | X | Seminal vesicle | X | Thyroids |
| | _____ | XX | Ovaries | X | Parathyroids |
| | RESPIRATORY | X | Oviduct | | |
| X | Trachea | X | Uterus | | _____ |
| X | Lung | X | Cervix | | OTHER |
| | Nose | | | X | Bone (femur, sternum) |
| | Pharynx | | | X | Skeletal muscle |
| | Larynx | | | X | Skin |
| X | Nasopharyngeal cavity* | | | X | All gross lesions and masses |
| | | | | X | Preputial gland |

* Tissues were preserved, but not examined microscopically.

## II. RESULTS

A. Observations :

1. Mortality - No treatment-related deaths occurred during the study. Two mice each in the control (1/sex) and 50-ppm (1/sex) groups and one 7000-ppm female were euthanized during weeks 11, 15, 24, 26, or 52. The death of an additional 7000-ppm female during week 4 was considered accidental. Two females each in the control and 7000-ppm groups were found dead; the control females were found dead during week 9 or 18 and the 7000-ppm females were found dead during week 13 or 16. All of the unscheduled deaths were considered to be incidental and not treatment-related.

2. Clinical signs - No treatment-related clinical signs of toxicity were noted in any treated group during the study.

B. Body weight and body weight gain - No treatment-related differences in body weights were observed in any treatment group relative to concurrent controls. Decreased body weights ($p \leq 0.05$ or 0.01) observed in the 7000-ppm males during 17/25 intervals (↓1-6%) and in the 7000-ppm females during 7/25 intervals (↓2-3%), were minor and considered not to be toxicologically important. Slightly increased body weights (↑2%, $p \leq 0.05$) in the 10-ppm males during week 17 were also considered not to be toxicologically important.

Overall mean body weight gain (as calculated by the reviewers) for the 7000-ppm males was lower (↓17%) than the controls (Table 2). Overall body weight gain for all other treatment groups was comparable to the respective control gains.

Table 2. Selected mean body weights (g) and overall mean body weight gain (g) in mice after treatment with ZA 1296 for 53 weeks.[a]

| Dose (ppm) | Week 1 | Week 3 | Week 6 | Week 10 | Week 17 | Week 29 | Week 41 | Week 53 | Overall BWG[c] Weeks 1-53 |
|---|---|---|---|---|---|---|---|---|---|
| **Males** | | | | | | | | | |
| 0[b] | 21.1 | 22.4 | 24.9 | 26.4 | 28.6 | 31.4 | 32.3 | 33.5 | 12.5 |
| 10 | 21.0 | 22.6 | 25.1 | 26.8 | 29.3* (↑2%) | 31.8 | 32.8 | 33.7 | 12.6 |
| 50 | 20.9 | 22.4 | 24.5 | 26.1 | 28.7 | 30.9 | 31.7 | 32.9 | 12.0 |
| 350 | 21.0 | 22.4 | 25.1 | 26.7 | 28.6 | 31.3 | 32.1 | 33.2 | 12.2 |
| 7000 | 21.1 | 22.1* (↓1%) | 24.4* (↓2%) | 25.6** (↓3%) | 28.0 | 29.9** (↓5%) | 30.4** (↓6%) | 31.6** (↓6%) | 10.4 (↓17%) |
| **Females** | | | | | | | | | |
| 0[b] | 17.1 | 18.3 | 21.3 | 23.1 | 24.6 | 26.3 | 26.7 | 27.8 | 10.7 |
| 10 | 17.1 | 18.4 | 21.5 | 23.0 | 25.0 | 26.1 | 27.1 | 27.8 | 10.8 |
| 50 | 17.2 | 18.1 | 21.1 | 22.8 | 24.6 | 26.1 | 26.9 | 28.3 | 11.1 |
| 350 | 17.0 | 18.4 | 21.1 | 22.9 | 24.8 | 26.2 | 27.0 | 27.7 | 10.7 |
| 7000 | 16.8 | 18.2 | 20.8* (↓2%) | 22.3** (↓3%) | 24.4 | 26.0 | 26.4 | 27.8 | 11.0 |

a Data obtained from study report Table 7, pages 61 through 68. Percent difference from controls is listed parenthetically. Adjusted (for initial body weight) mean data were used.
b Data from two control groups were averaged by the reviewer.
c Overall body weight gains were calculated by the reviewer.
* Statistically different from pooled controls at $p \leq 0.05$.
** Statistically different from pooled controls at $p \leq 0.01$.

C. Food and compound intake/water consumption

1. Food consumption and utilization - No treatment-related differences in food consumption were observed in any treatment group. Increased ($p \leq 0.05$ or 0.01) food consumption was observed in the 7000-males (↑9%) at week 40, in the 350-ppm males (↑2-11%) during 13/23 intervals, and in the 7000- (↑5-15%), 350- (↑14%), 50- (↑7-8%), and 10- (↑10-12%) ppm females during 1-3/23 intervals. The increases in food consumption were not dose-dependent, and therefore, considered unrelated to treatment. Food utilization (measured during weeks 1-13 only) was reduced in the 7000-ppm males and females



9

261

during weeks 1-4 (↓16-23%, p≤0.05 or 0.01). Overall food utilization (weeks 1-13) was slightly decreased in the 7000-ppm males (↓9%, p≤0.05).

2. Compound consumption - The achieved mean dosages based on nominal dietary concentrations, actual body weights, and actual food consumption are shown in Table 1.

3. Water consumption - Water consumption was not measured.

D. Ophthalmoscopic examination - No treatment-related ocular effects were observed.

E. Blood analyses

1. Hematology - No treatment-related differences in hematology parameters were observed during this study. All male treated groups displayed dose-dependent decreases (p≤0.05 or 0.01) in white blood cell (↓34-45%) and lymphocyte (↓36-47%) counts at week 14 compared to concurrent controls. These decreases were not sustained over time, and therefore, were considered not to be treatment-related. All male treated groups exhibited decreased (p≤0.05 or 0.01) monocyte counts (↓38-61%) and eosinophil counts (↓39-57%) at week 14; however, these decreases were neither dose-dependent nor sustained over time, and considered not to be treatment-related. At week 53, decreased white blood cell, lymphocyte, and large unsustained cell counts (↓47, 53, and 59%, respectively; p≤0.05) in the 7000-ppm females lacked corroborating evidence of toxicity, and therefore were considered unrelated to treatment. At week 53, decreased monocyte and eosinophil counts (↓34 and 37%, respectively; p≤0.05) in the 7000-ppm males were not sustained over time (not observed at week 27), and therefore were considered unrelated to treatment. Differences (p≤0.05) observed at week 14 that were neither dose-related nor sustained over time, and therefore, considered unrelated to treatment were as follows: (i) decreased neutrophil count (↓47%) in the 350-ppm males; (ii) decreased eosinophil count (↓69%) in the 350-ppm females; and (iii) increased large unstained cell count (↑163%) in the 10-ppm females. Decreased basophil count (↓100%, p≤0.01) in the 50-ppm females at week 27 was neither dose-related nor sustained over time and therefore, was considered not to be treatment-related. Decreased neutrophil count (↓32%, p≤0.05) in the 50-ppm females at week 53 was not dose-related and therefore, considered unrelated to treatment. Minor, sporadic differences (↓4-↑5%, p≤0.05 or 0.01) in various hematological parameters observed in both sexes at various points throughout the study were considered not to be treatment-related.

2. Clinical chemistry - No treatment-related differences in clinical chemistry parameters were observed during this study. Differences that were statistically significant (p≤0.05 or 0.01) at week 27, but were neither dose-dependent nor sustained over time included the following: (i) increased total protein (↑7%) in the 50-ppm males; (ii) decreased total bilirubin (↓30%) in the 350-ppm females; (iii) increased creatine kinase (↑168%) in the 50-ppm females; (iv) increased aspartate aminotransferase (↑48%) in the 50-ppm females; and (iv) slightly decreased sodium (↓1%) in the 10-ppm females. Clinical chemistry

parameters that were statistically significant ($p \leq 0.05$ or 0.01) at week 53 but were not dose-dependent and considered unrelated to treatment included the following: (i) decreased total bilirubin (↓21%) in the 350-ppm males; (ii) increased aspartate aminotransferase (↑40%) in the 50-ppm males; (iii) increased phosphorus in the 50-ppm males and in the 50- and 10-ppm females (↑17%, each); and (iv) increased potassium in the 10-ppm females (↑21%). Increased ($p \leq 0.05$) creatine kinase in the 7000-ppm males (↑195%) and 50-ppm females (↑168%) at week 27 was due to high values for several animals in each group. When these high values were excluded from analysis, a treatment-related effect was not observed . At week 53 in the 7000-ppm animals, decreased ($p \leq 0.05$ or 0.01) creatinine was observed in the males (↓5%), and decreased plasma urea (↓26%) and alkaline phosphatase (↓27%) were observed in the females; however, in the absence of corroborating histopathological evidence of toxicity and since the values were within normal reference ranges, these findings have no toxicological significance. Decreased creatinine levels ($p \leq 0.05$ or 0.01) in the 7000-, 350-, and 50-ppm females (↓13, 11, and 6%, respectively) at week 53 were dose-related; however, these differences were relatively minor compared to controls, and there was no corroborating toxicological evidence to suggest that they were treatment-related.

F. Urinalysis - No treatment-related differences in urinalysis parameters were observed in any treatment group. Minor differences ($p \leq 0.05$ or 0.01) considered not to be toxicologically important and unrelated to treatment included increased urine specific gravity (↑1%) in the 7000-ppm males and females and the 350-ppm females at various time points throughout the study, and decreased urine pH in the 7000-ppm males at weeks 13, 26, and 52 (6.3-6.7 treated vs. 6.9-7.0 control), in the 350-ppm males during week 52 (6.5 treated vs. 7.0 control), and in the 7000- and 350-ppm females during weeks 13 and 26 (6.8-6.9 treated vs. 7.1-7.3 control). At week 52, increases ($p \leq 0.01$) observed in urine volume (↑89%) in the 50-ppm females and urine pH (7.5 treated vs. 7.0 for control) in the 10-ppm females were not dose-related and therefore, were considered not to be treatment-related. Dose-related increases in ketones were observed in urine from all treated groups (qualitative analysis). This effect may be related to the urinary excretion of the metabolites of the test compound; however, the lack of corroborating toxicological evidence suggests that this finding is not of biological relevance.

G. Sacrifice and Pathology:

1. Organ weight - No treatment-related differences in absolute or relative organ weights were observed. Decreased absolute brain weights (↓4%, $p \leq 0.05$) in the 7000- and 350-ppm males compared to concurrent controls were likely due to the slightly reduced mean final body weights for both groups compared to the controls. A minor decrease in brain weight (adjusted for body weight) in the 50-ppm males (↓4%, $p \leq 0.05$) was minor, not dose-related, and considered unrelated to treatment. In the 7000-ppm females, increases (↑7%, $p \leq 0.01$) in kidney weight (adjusted for body weight) and liver weight (adjusted for body weight) were minor and considered unrelated to treatment.

2. Gross pathology - No test substance-related findings in gross pathology were observed.



//

3. Microscopic pathology - No test substance-related findings in microscopic pathology were observed. At week 53, an increased incidence of eosinophilic change of the gallbladder epithelium was observed in 5/10 of the 7000-ppm females (vs. 0/20 controls). This finding was not accompanied by microscopic changes in the liver and it was stated that it is a common finding in older mice.

## III. DISCUSSION

A. Investigator's conclusions - Oral administration of mesotrione (ZA 1296) for 53 weeks was systemically toxic at 7000 ppm. The test compound at this dose caused reduced body weights and food utilization in both sexes, adrenal gland changes in males, and gallbladder changes in females. The NOAEL for this study was 350 ppm.

B. Reviewer's discussion - In this chronic oral toxicity study, mesotrione (ZA 1296) was administered continuously in the diet to 60 C57BL/10J$_f$CD-1 mice/sex/dose at nominal dose levels of 10, 50, 350, or 7000 ppm (equivalent to [M/F] 1.5/2.1 7.8/10.3, 56.2/72.4, and 1114.0/1494.5 mg/kg/day, respectively) for 3, 6, or 12 months. Two control groups of 60 C57BL/10J$_f$CD-1 mice/sex (120/sex) were fed untreated diet for 3, 6, or 12 months. There were 20 treated mice/sex/group per time point. Analysis of the test substance indicated that the mixing procedure was adequate and that the variation between nominal and actual dosage to the study animals was acceptable. Furthermore, the results confirmed the stability of the test substance at room temperature for a period of 16 days.

No treatment-related mortality was observed in any treated group. Two mice each in the control (1/sex) and 50-ppm (1/sex) groups and one 7000-ppm female were euthanized during weeks 11, 15, 24, 26, or 52. The death of an additional 7000-ppm female during week 4 was considered accidental. Two females each in the control and 7000-ppm groups were found dead during weeks 9, 13, 16, and 18. All of the unscheduled deaths were considered to be incidental and not treatment-related. Clinical signs, body weights, food consumption, hematology, clinical chemistry, and urinalysis parameters, ophthalmoscopic observations, organ weights, and gross and histopathological findings were unaffected by the test substance.

Body weight gain in the 7000-ppm males was reduced overall by 17%. Reductions in body weight gain appeared to occur throughout the study. In conjunction with a 23% decrease in food utilization during the first 4 weeks of the study, an overall decrease in food utilization of 9% from weeks 1-13, and no decrease in food consumption throughout the study, the evidence suggests that there was a toxicologically significant effect in the 7000-ppm males.

Under the conditions of this study, there was no evidence of carcinogenic potential.

**The LOAEL for this study is 7000 ppm (equivalent to 1114.0 mg/kg/day in males and 1494.5 mg/kg/day in females) based on decreases in body weight gain and food**



12

**utilization in males. The NOAEL is 350 ppm (equivalent to 56.2 mg/kg/day in males and 72.4 mg/kg/day in females).**

The submitted study is classified as **acceptable/guideline (§83-1b)** and satisfies the guideline requirements for a chronic oral toxicity study in rodents.

C. Study deficiencies - The following deficiencies were noted, but will not change the conclusions of this review. First, not all of the organs and tissues required by §83-1 were collected and weighed. The spleen, heart, thymus and epididymides should have been weighed. A measure of blood clotting potential, such as prothrombin time or activated partial thromboplastin time, should have been performed. Lastly, the temperature and relative humidity of the animal room should have been maintained at 22 ± 3 °C and 50 ± 20%, respectively.