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

MEMORANDUM

TO: Tim Gardner, PM-17
Registration Division (TS-767)

THRU: Robert B. Jaeger, Section Head
Review Section #1
Toxicology Branch/HED (TS-769)

SUBJECT: Diflubenzuron 37100-8: Registrant's Request for Removing some Precautionary Label Statements and Review of the Lifetime Oncogenic Study of Diflubenzuron in Rats (52-Week Interim Report) Accession No. 250974, 250975, 250976, 250977. Caswell No. 346A.

Recommendation:

1. Based on the acute oral and dermal toxicity data of Dimilin W-25 previously submitted, Toxicology Branch agrees with the Registrant that these data demonstrate a low order of acute toxicity (e.g., Tox. Cat. IV). However, the EPA Decision Document (March 26, 1979) addressed three options for the "conditional use" of Dimilin and it was stated by EPA that "Diflubenzuron would be classified for restricted use and would be applied only by certified applicators. These applicators would be required to wear protective clothing and respirators." These requirements were based on the "uncertainties" raised by long-term feeding studies. Additional studies were required and at present TB has only received interim reports. TB, therefore, believes that subject labeling should not be deleted until all the toxicity data requested in 1979 have been received and reviewed.

2. Toxicology Branch has reviewed the 52-week interim oncogenic report in rats, which contains insufficient data to assess the oncogenic potential related to the treatment of Dimilin in Sprague-Dawley rats.

1979

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...oject No. 553-122,
...eek Interim Report

...

...the oncogenic potential of
...when administered in the feed
...eeks of a lifetime study is
...ubdivision F Hazard Evaluation
...the following specific test

1. Test

The ... of 600 CR-CD Sprague-Dawley rats (30... ...females). The rats were acclimated ... for approximately four weeks, then ... ed ... 4 dosage levels of diflubenzuron in ... (156, 625, 2500, and 10,000 ppm) plus a control group. There were 50 males and 50 females in each treated group and 100 males and 100 females in the control group.

2. Preparation of the Test Diet

An appropriate amount of the test compound was premixed with a portion of the basal diet of Purina rodent laboratory chow. The premix was then added to the correct amount of commercial feed to attain the proper dosage levels and again mixed. Diets of 156, 625, 2500, and 10,000 ppm of diflubenzuron were analyzed during the first 12 weeks of study and approximately every 4 weeks thereafter.

3. Observation and Examination

All animals were observed for mortality and moribundity. Individual body weights, food consumption, and clinical signs were examined weekly throughout the test period for all groups of animals. Hematology studies were determined for the first ten rats/sex/group at week 52. Blood samples of rats were collected during the week 42 and 44 of this study for serology screening study. Necropsy was performed for all animals that were found dead during the study. The external surface, all orifices, cranial cavity, carcass, external and cut surfaces of the brain and spinal cord, nasal cavity and paranasal sinuses, thoracic, abdominal, and pelvic cavities, and their

viscera, cervical tissues and organs, middle ear, and zymbal's gland. The following tissues from each animal found dead were collected and stained with hematoxylin and eosin for microscopic examinations: nervous, special sense, endocrine, urogenital, digestive, lymphatic, hemic, cardiovascular, respiratory, and musculoskeletal systems. For each sacrificed animal, the brain, heart, liver, kidneys, spleen, testes with epididymides were weighed.

4. Statistical Analysis

Data were analyzed statistically using one-way classification analysis of covariance (Snedecor and Cochran, 1967). If significant effects were indicated, the Games and Howell's modification of Tukey-Kramer's significant difference test was used for comparing the group means between the treated and control group. All analyses were evaluated at the 5% probability (one-tailed).

Results:

1. Mortality

No significant treatment-related effect on the incidence of mortality was observed during the first 52 weeks of this study.

2. Clinical Examinations

Swollen neck, lacrimation, weight loss, rhinorrhea, blood crust on eyelids, and swollen ear were observed in the control and treated animals at the week 40. These incidental clinical signs were not related to the treatment of diflubenzuron.

3. Body Weights

A lower mean body weight value was consistently observed in the 625, 2500, and 10,000 ppm level groups when compared to the respective control value throughout the first 52-weeks of study. But no significant differences were noted for the treated animals in analysis of growth rates through week 52 of study.

4. Food Consumption

There were no significant differences in the mean weekly food consumption values between the treated

and control during the period of study. 003607

5. Hemotology

(a) Mean Clinical Hematology Values of Male Rats at the Week 52

Dose (ppm)	0	156	625	2500	10,000
*HCT (%)	47.9	47.3	44.1	45.5	42.5*
*HGB (g/dl)	16.5	15.7	14.6*	15.6	15.2*
*RBC (Mi/ul)	8.7	8.3	7.6*	8.0	7.4*
WBC (Th/ul)	9.2	10.2	11.6	11.1	11.0
*MCH (Pg)	18.9	18.9	19.3	19.5	20.5*
MCHC (%)	34.4	33.2	33.3	34.4	35.8
*RETIC (%RBC)	0.9	0.8	1.6	1.7	2.2*
*Metheme (g/dl)	0.04	0.31*	0.22*	0.37*	0.38*
*Sulfheme (g/dl)	0.02	0.21*	0.14*	0.11*	0.16*

* Significant changes; HCT-Hematocrit; HGB-Hemoglobin; RBC-Red Blood Cell; WBC-White Blood Cell; MCH-Mean Corpuscular hemoglobin; RETIC-Reticulocytes Count; Metheme-Methemoglobin; Sulheme-Sulfhemoglobin.

Summary of hematological findings for male rats:

Significant decreases in the mean hematocrit values were noted in the 10,000 ppm level group. The mean hemoglobin and erythrocyte values were also significantly decreased for the groups treated with 625 and 10,000 ppm diflubenzuron. The mean corpuscular hemoglobin and reticulocyte counts were significantly increased in the high dose level groups. Methemoglobin and sulfhemoglobin were also significantly increased in all of the groups treated with diflubenzuron.

(b) Mean Clinical Hematology Values of Female Rats at the Week 52

Dose (ppm)	0	156	625	2500	10000
*HCT (%)	46.4	45.5	45.4	43.0	41.0*
*HGB (g/dl)	15.4	15.0	14.4	13.6*	13.4*
*RBC (Mi/ul)	8.1	7.8	7.6	7.1*	6.5*
*WBC (Th/ul)	6.8	7.2	5.9	7.1*	7.5*
*MCV (fl)	57.3	58.1	60.1	60.9*	62.9*
*MCH (Pg)	19.1	19.2	19.0	19.3	20.6
*MCHC (%)	33.3	33.1	31.7*	31.7*	32.7
Platelet (Th/ul)	941.0	930.0	926.0	990.0	1063.0
*RETIC (% RBC)	1.0	1.10	1.8	3.2*	5.1*
*Metheme (G/dl)	0.1	0.1	0.2*	0.3*	0.3*
*Sulfheme (g/dl)	0.02	0.03	0.14*	0.02	0.05

* - Significant changes; MCV - Mean Corpuscular Volume MCHC - Mean Corpuscular Hemoglobin Conc.; Metheme - Methemoglobin; Sulfheme - Sulfhemoglobin.

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Summary of hematological findings for females rats:

The mean hematocrit values for the 10,000 ppm level group were significantly lower than the control value. The mean hemoglobin and erythrocyte values were significantly decreased for the groups treated with 2500 and 10,000 ppm diflubenzuron. There were also decreases in the mean corpuscular hemoglobin concentration in the 625, 2500, and 10,000 ppm level groups. The mean corpuscular volume and reticulocyte counts were significantly increased in the groups treated with 625, 2500, and 10,000 ppm diflubenzuron. The sulfhemoglobin value was significantly increased in 625 ppm level group only.

6. Serology Screening for the Presence of Microbes

The presence of sendai virus and sialodacryoadenitis was detected in serum samples tested by the hemagglutination inhibition and enzyme-linked immunosorbent assay techiques.

7. Gross Pathology Findings for Animals Found Dead or Sacrificed In Extremis during the Weeks 1-52.

	Male					Female				
Dose Levels (PPM)	0	156	625	2500	10,000	0	156	625	2500	10,000
No. Examined	4	1	0	2	4	0	1	1	3	0
Brain										
identation	0	0	-	0	0	-	0	1	1	-
Meniinges Black	0	0	-	0	1	-	1	0	0	-
Pituitary										
Enlarged	0	0	-	0	0	-	0	0	2	-
Lung										
Mottled	2	1	-	0	1	-	0	0	0	-
Darkened	0	0	-	1	2	-	1	0	0	-
Liver										
Thickened	0	0	-	0	1	-	0	0	0	-
Enlarged	0	0	-	1	1	-	0	0	0	-
Darkened	0	0	-	0	1	-	0	0	0	-
Heart										
Enlarged	0	1	-	0	0	-	0	0	0	-
Spleen										
Enlarged, Red	0	0	-	1	1	-	0	0	0	-
Kidney(s)										
Modulla Darkened	0	1	-	0	1	-	0	0	1	-
Cortex Dark	0	1	-	0	1	-	0	0	0	-
Adrenal(S)										
Enlarged, Darkened	0	0	-	0	0	-	0	0	2	-
Stomach										
Dark Lining	0	0	-	1	0	-	0	0	1	-
Grandular Mucosa	0	1	-	0	0	-	1	0	0	-
Intestines										
With gas	0	1	-	0	0	-	0	0	0	-
Lymph Nodes										
Mesenteric	0	0	-	0	1	-	0	0	0	-
Mandibular Red	0	0	-	1	1	-	0	0	0	-
Uninary Bladder										
Dark red fluid	0	0	-	1	1	-	1	0	1	-
Seminary Vescles	0	1	-	0	0	-	-	-	-	-
Ovary	-	-	-	-	-	-	0	0	1	-

Summary of Necropsy Findings: The total number of animals examined for each treated and control group was inadeqate to draw any meaningful conclusion.

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8. Individual Histopathological Findings Related to the Neoplastic Lesions for Animals Found Dead or Sacrificed In Extremis During the Weeks 1-52.

	Male					Female				
Dose Levels (PPM)	0	156	625	2500	10,000	0	156	625	2500	10,000
Pituitary										
No. Examined	4	1	0	2	4	0	1	1	3	0
No. of M-carcinoma	0	0	0	0	0	0	0	1*	0	0
Lung w/Bronchi										
No. Examined	4	1	0	2	4	0	1	1	3	0
No. of X-Monocytic Leukemia	0	0	0	0	1*	0	0	0	0	0
Skin										
No. Examined	4	1	0	2	4	0	1	1	3	0
No. of M-Hemangio-Sarcoma	0	0	0	0	1*	0	0	0	0	0
Tissue Mass(es)										
No. Examined	1	0	0	0	0	0	0	0	0	0
No. of M-Lipocarcoma	1*	0	0	0	0	0	0	0	0	0

Summary of Histopathological Findings: Because the tumor profile of untreated male and female Sprague-Dawley rats was not given in the report, the incidences of tumor formation including the carcinoma of pituitary, disseminated monocytic leukemia and hemangiosarcoma of the skin found in the test animals are considered inconclusive in this 52-week Interim Report.

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Conclusion:

1. Sixteen out of 600 Sprague-Dawley rats died during the first 52 weeks of study. The cause of death was apparently due to the microbial infection and was not related to the treatment of diflubenzuron.

2. The evidence of tumorgenicity related to the treatment with diflubenzuron in rat is incomplete at this stage of study. Further examination of the development of tumor types identified must be carefully pursued in the final stage of this study.

3. Classification: Study Incomplete - Interim Report.

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DCR-44961:Chen:2/7/84/Del.2/19/84:mar
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