12/4/85

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

004823

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: WEEDONE 2,4-DP Woody Plant Herbicide
Evaluation of a 13-Week Rat Clinical
Chemistry Study Submitted as a
Supplement for the Completion of
the Chronic Feeding Study
EPA Registration No. 264-231

Accession No. 250351
Tox Chem #320

FROM: William B. Greear, M.P.H. William B. Greear 11/25/85
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TO: Richard Mountfort, PM Team #23
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THRU: Albin B. Kocialski, Ph.D.
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and

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In a letter dated May 9, 1983, W.A. Davis of Union Carbide Agricultural Products Company, Inc. submitted the results of a 13-week subchronic feeding study in rats. In his letter, W.A. Davis states that because the previously submitted chronic rat study was deficient, i.e., it lacked appropriate "hematological evaluations," it was "agreed that a 13-week subchronic study that addressed the area of blood chemistry would be an adequate supplement to validate the base chronic report." The study has been evaluated and could support the classification of Supplementary Data.

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DATA EVALUATION RECORD

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Subject: A365 (2,4-DP Acid): 24-Month Oral Chronic Toxicity Study in Rats (13-Week Rat Clinical Chemistry Supplementary Study)

Chemical: 2,4-DP acid (95% Technical)

Accession No.: 250351

EPA Registration No.: 264-231

Laboratory: Not reported

Study No./Report Date: Study number not reported/December 14, 1981

Study Authors: K. Mitsumori, T. Saito, T. Miyaoka, Y. Shirasu

Test Material/Purity: 2-(2,4-dichlorophenoxy)propanoic acid (95% pure)

Testing Period: July 19, 1981 to September 24, 1981

Classification: Supplementary Data

Materials and Methods:

Animals

Four-week-old specific pathogen free Fischer CDF (F-344) rats were obtained from Charles River Co., Ltd. (The location of the breeding facility was not reported.) The animals were acclimated to the laboratory for 1 week prior to initiation of the test. The animals were assigned to groups as indicated below. (No indication was given that a randomization procedure was used to distribute the animals.)

Treatment	Male Rats	Female Rats
Control	10	10
100 ppm	10	10
300 ppm	10	10
1000 ppm	10	10
3000 ppm	10	10

Environment

Animals were housed in wire-mesh stainless steel cages in groups of five in a barrier-sustained animal room at a temperature of 24 ± 1 °C and a relative humidity of 55 ± 5 percent. The animal room received illumination for 14 hours/day and the room received 12 changes of air per hour. Food (Oriental M

powdered food) and water were available ad libitum. Test diets were prepared at the treatment levels indicated above. (There was no indication that the test diets were analyzed for concentration, stability, or homogeneity.)

Observations and Measurements

Animals were observed daily for toxic signs and individual body weights were recorded weekly. After 84 days of treatment, urine samples were obtained from all animals. The urine was examined for specific gravity, pH, protein, glucose, ketones, occult blood, and urobilinogen. On day 91, blood samples were obtained from all animals after the animals had been lightly anesthetized with ether. The hematologic parameters examined included hematocrit, hemoglobin, erythrocyte count, erythrocyte indices (MCV, MCH, MCHC), platelet count, leukocyte count, differential leukocyte count, and reticulocyte count. The following clinical chemistry determinations were made on serum samples of blood obtained for hematologic examination: total protein, albumin, globulin, albumin/globulin ratio, alkaline phosphatase, lactic dehydrogenase, glucose, blood urea nitrogen, glutamic oxaloacetic transaminase, glutamic pyruvic transaminase, gamma-glutamyl transpeptidase, total bilirubin, direct bilirubin, total cholesterol, calcium, sodium, and potassium.

All animals were subjected to a gross necropsy examination. The following organs from animals sacrificed at termination were weighed: brain, pituitary, thyroids with parathyroids, heart, thymus, liver, kidneys, spleen, adrenals, gonads, and skeletal muscle (M. triceps surae of unilateral hind leg). The following tissues from all animals sacrificed in extremis and at termination were preserved in 10 percent neutral buffered formalin: brain, spinal cord (cervical, thoracic, and lumbar regions), sciatic nerve, liver, pancreas, salivary glands, tongue, esophagus, stomach, duodenum, jejunum, ileum, cecum, colon, larynx, trachea, lung, heart, thoracic aorta, spleen, thymus, lymph nodes (cervical, mesenteric), bone with marrow (sternal, femoral), tibio-femoral joint, skeletal muscle (M. triceps surae), kidneys, urinary bladder, testes, epididymides, seminal vesicles, coagulating glands, prostate, mammary gland (abdominal region), ovaries, uterus, pituitary, thyroids, parathyroids, adrenals, skin (lumbodorsal region), eyes, intra-orbital lacrimal glands, head including nasal cavity with paranasal sinuses and middle ear, and any gross abnormalities.

Statistics

Analysis of variance followed by Student's t-test was used to determine the significance of the results.

Results:

Clinical Signs/Body Weight

No abnormal clinical signs were noted during the study. One female in the 100 ppm group was sacrificed in extremis on day 38 due to the presence of a spontaneous tumor (not identified). Body weight gain was decreased in males and females in the 3000 ppm group.

Hematology

The hematocrit, hemoglobin concentration, and erythrocyte count were decreased in females in the 1000 and 3000 ppm group and in males in the 3000 ppm group. Males in the 3000 ppm group had slight increases in the reticulocyte count, MCV, MCH, and MCHC. Females in the 3000 ppm group had a slight increase in MCV and slight decrease in MCHC.

Clinical Chemistry

Males in the 3000 ppm group had increases in alkaline phosphatase, total bilirubin, albumin and albumin/globulin ratio, and decreases in glutamic pyruvic transaminase, total protein, globulin, total cholesterol, and calcium. Sodium and potassium values were comparable to controls. Females in the 3000 ppm group had increases in total bilirubin, albumin and albumin/globulin ratio, and decreases in glutamic oxaloacetic transaminase, glutamic pyruvic transaminase, gamma-glutamyl transpeptidase, total protein, globulin, total cholesterol, and calcium. Sodium and potassium values were similar to control values. Males in the 1000 ppm group had increases in alkaline phosphatase, total bilirubin, albumin and albumin/globulin ratio, and a decrease in globulin. Females in the 1000 ppm group exhibited an increase in albumin/globulin ratio, and a decrease in globulin.

Urinalysis

No significant effects were observed; however, there was a decrease in the number of females in the 3000 ppm group in which ketone was detected in the urine.

Necropsy

Three males in the 3000 ppm group had dark-colored livers. A fourth male in this group had a liver with a coarse surface. No other abnormalities observed could be related to treatment.

Organ Weight

(Absolute): Males in the 3000 ppm group exhibited a decrease in the weight of the heart, spleen, and muscle (M. triceps surae of unilateral hind leg); and increase in liver weight. Females in the 3000 ppm group had decreases in the weight of the heart, spleen, thymus, and muscle (M. triceps surae of unilateral hind leg). Males in the 100, 300, and 1000 ppm groups had increases in kidney weight. Females in the 1000 ppm group also had a decrease in spleen weights.

(Relative): Male relative kidney weight was increased in all test groups when compared to the controls, however, the increase did not appear to be dose-related. Males in the 3000 ppm group had increases in the relative weight of the brain and liver. Females in the 3000 ppm group had increases in the relative weight of the brain, liver, thyroid, and kidney. The relative weight of the kidney was also increased in females in the 1000 ppm group.

Discussion and Conclusions:

No abnormal clinical signs of toxicity were observed during the course of the study. Body weight gain was decreased in males and females in the 3000 ppm group. Males and females in the 3000 ppm group and females in the 1000 ppm group had decreases in hematocrit, hemoglobin concentration, and erythrocyte count indicating the probable development of anemia. Males in the 3000 ppm group also had a nonsignificant increase in the reticulocyte count, and increases in MCV, MCH, and MCHC. Females in the 3000 ppm group had a slight increase in MCV and slight decrease in MCHC. At doses of 1000 ppm or higher, males displayed increases in alkaline phosphatase, total bilirubin, albumin and albumin/globulin ratio, and decreases in globulin. Males in the 3000 ppm group also had decreases in total protein, total cholesterol, and glutamic pyruvic transaminase. Females in the 3000 ppm group had increases in total bilirubin, albumin and albumin/globulin ratio, and decreases in gammaglutamyl transpeptidase, glutamic pyruvic transaminase, glutamic oxaloacetic transaminase, total protein, globulin, blood urea nitrogen, total cholesterol, and calcium. Females in the 1000 ppm group had an increase in the albumin/globulin ratio, and a decrease in globulin. Relative and absolute organ weights varied considerably. The absolute and relative weight of the liver was increased in males in the 3000 ppm group, and the relative weight of the liver was increased in females in the 3000 ppm group. At gross necropsy, three males in the 3000 ppm group had dark livers. An additional male in the high-dose group had a liver with a coarse surface. This information indicates a probable effect on the liver at the 3000 ppm level.

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LEL = 1000 ppm (based on decreases in hematocrit, hemoglobin, erythrocyte count, and globulin and an increase in albumin/globulin ratio in females; and on an increase in alkaline phosphatase, total bilirubin, albumin and albumin/globulin ratio, and a decrease in globulin in males).

NOEL = 300 ppm.

Classification: Supplementary Data. The study is acceptable and fulfills the clinical chemistry aspects of the chronic study.

(Note: The purpose of the study is to provide data on blood chemistry and hematology, since the chronic rat study failed to include these data - see letter from W.A. Davis, Union Carbide, dated May 9, 1983, EPA Correspondence No. 124-83.

Recommendations:

It is recommended that:

- The sponsor provide the identity of the testing laboratory for this study.
- Provide the study report number.
- Provide the source of the Fischer CDF (F-344 rats).
- Provide a statement with regard to the randomization procedure.
- Provide a statement with regard to the concentration, stability, and homogeneity of the material in feed.