10-10-91 008711

Primary Reviewer: David S. Liem, PhD David S. Liem 10/8/91
Tox. Branch II/Section II/HED (H7509C)
Secondary Reviewer: Clark Swentzel, Section Head K. Clark Swentzel 10/10/91
Tox. Branch II/Section II/HED (H7509C)

DATA EVALUATION REPORT

STUDY TYPE: Rangefinding Oral Toxicity Study with Sulfluramid (20% linear/80% branched isomers) - Non-Guideline Study

MRID NO.: 418699-01 CASWELL NO: 454E HED PROJECT NO: 1-1257

TEST MATERIAL: Sulfluramid (20% linear/80% branched isomers)
MRD 89-513

SYNONYMS: N-ethyl perfluoro-octanesulfonamide

DOSAGES: 0, 30, 60, 120, 240, and 480 ppm

SPONSOR: Griffin Corporation, Rocky Ford Rd., Valdosia, GA 31603

TESTING FACILITY: Exxon Biomedical Sciences, Inc., Mettelers Rd., CN 2340, East Millstone, NJ 08875

TITLE OF REPORT: Rangefinding Study in Rats with Sulfluramid(20/80)

AUTHOR(S): Roger T. Keefe STUDY NUMBER: 246754

REPORT ISSUED: November 16, 1990

CONCLUSIONS:

Six groups of five males and five females of Sprague-Dawley rats were fed with 0, 30, 60, 120, 240, and 480 ppm sulfluramid 20/80 in the diet for a period of 28 days.

A number of liver abnormalities were noted and it appears that the liver is the target organ for "sulfluramid 20/80".

Based on mortality, body weight, food consumption, and clinical signs, treatment-related effects were observed in the 240 and 480 ppm of both sexes as well as in the 120 ppm females. The data also show that females appeared to be more sensitive than the males.

The definitive NOEL and LOEL can not be determined because not all 30, 60, 120 ppm livers were microscopically evaluated, and also because of the small number of animals used in the study. Based on body weight and food consumption data, the LOEL is determined to be 240 ppm for the males and 120 ppm for the females. Accordingly, the NOEL is 120 ppm for the males and 60 ppm for the females.

There is some concern that livers of the intermediate dose groups (30, 60, and 120 ppm) were not histopathologically evaluated.

The test article analysis showed that it contained a 31.7% linear and a 62.6% branched Sulfluramid isomers. This mixture deviated somewhat from the intended 20% linear and 80% branched isomers mixture indicated in the study report.

The investigators should clarify why some livers of the intermediate dose groups were not evaluated and also why the linear and branched Sulfluramid isomer ratio based on the analysis deviated from the intended isomer ratio.

Title of Report: Rangefinding Study in Rats with Sulfluramid(20/80)

Author: Roger T. Keefe Report Date: November 16, 1990

Study Period: September 21 - October 19, 1989 (In-life)

Study Number: 246754 MRID NO.: 418699-00

Test Material: Sulfluramid (20% linear/80% branched isomers)

Test Animal: Crl:CD(BR) (Sprague Dawley) Rats

A. OBJECTIVES: The objectives of this study were to evaluate the oral toxicity of sulfluramid (20/80) (MRD-89-513) and to determine dose levels for a longer term study in rat.

B. MATERIALS:

1. Test compound: Sulfluramid (20/80). Description - Pale yellow paste (31.7% linear and 62.6% branched isomers). Batch # - AN-90145. The purity was not reported.

2. Test animals: Species: Crl:CDBR (Sprague-Dawley) Rats, Source: Charles River Breeding Laboratories, Kingston, NY; Age: Approximately 7 weeks at start of study; Body weights were 227.1-262.6 g (males) and 160.8-206.1 g (females); Acclimation: 14 days; Individually caged.

C. STUDY DESIGN:

1. Animal Assignment

Rats were assigned to the following test groups:

DOSE GROUPS	DOSE (ppm)	TOTAL MALES	TOTAL FEMALES
Control	0	5	5
Group 1	30	5	5
Group 2	60	5	5
Group 3	120	5	5
Group 4	240	5	5
Group 5	480	5	5

2. Dosing Duration

Treated rats were fed with sulfluramid in the diet for a period of 28 days.

3. Diet preparation

Diet was prepared by mixing the test material dissolved in methanol. The methanol was removed by evaporation. The dried rat diet containing the test material was added to a larger quantity of the basal diet. The feed mixtures were prepared twice during the study. Half of each batch was fed the first week and the other half was stored frozen and fed during the second week after mixing. It was not noted that samples of the diet mixtures were analyzed for stability and concentration. Rats received feed and water ad libitum.

4. Environmental Parameters

Temperature: 68-76°F; Relative Humidity: 40-70%; Photoperiod: Approximately 12 hrs light/dark cycle.

D. EXPERIMENTAL EVALUATIONS

Clinical Observations

All rats were checked for signs of toxicity twice daily, and once daily on weekends.

Individual Body Weights and Food Consumption

Individual body weights were recorded during the week prior to study initiation, on days 0, 7, 14, 21, and 28. Body weights were also recorded for moribund sacrificed or rats found dead. Food consumption were recorded weekly.

Terminal Sacrifice

On day 28 of study, all surviving rats were sacrificed using CO_2 asphyxiation followed by exsanguination. All rats were subjected to gross macroscopic examination, including physical examination of external surfaces, all orifices, cranial, spinal, neck, nasal/perinasal, thoracic, abdominal and pelvic cavities with their associated organs and tissues. The testes and epididymides from all surviving males were weighed.

c. Histopathological Evaluations

At terminal necropsy, the testes and epididymides from all surviving males were harvested, fixed in Bouin's fixative, then they were processed, sectioned, and stained with hematoxylin and eosin for histopathological evaluations. Grossly abnormal liver of the 120 and 240 ppm dose groups were also harvested, fixed, processed and evaluated histopathologically. All other gross lesions were fixed in 10% neutral buffered formalin for possible future histologic evaluation.

d. Statistical Analysis of the Data

The means and standard deviations of organ weights, organ/body weight ratios, body weights, and food consumption were computed. Statistical tests were not performed in this study.

E. A signed Statement of Confidentially Claim, a Statement of Compliance with EPA's FIFRA GLP, and a Quality Assurance Statement were provided in the study report.

F. RESULTS AND DISCUSSIONS:

The study report did not clearly indicate that the control group used in this study (Sulfluramid 20/80) also served as the control for another study (Sulfluramid 70/30). This is confirmed by comparing the data of the controls of the four Sulfluramid rangefinding studies (Sulfluramid 20/80, 70/30, 90/10, and 99+).

On p. 8 of the study report it was noted that the certificate of Analysis described the test article as a 31.7% linear and 62.6% branched Sulfluramid isomers. The isomer ratio was different than the intended test article of 20% linear and 80% branched isomers mixture.

1. Mortality and Clinical Observations

Mortality and pertinent clinical observations are summarized in the attached Appendix A.

a. Mortality

Mortality only occurred in the 240 and 480 ppm dose groups. Two males and all females of the 240 ppm, and two females of the 480 ppm dose groups survived until study termination. Deaths in the 480 ppm group occurred on days 10, 18 (2 rats), 22, and 29 for males, and on days 27, 28, and 29 for the females. Deaths in the 240 ppm group occurred on days 25, and 29 (2 rats) for males.

b. Clinical Observations

Clinical signs of toxicity were generally confined to the 240 and 480 ppm dose groups. Hyperactivity was observed in one 240 ppm and in all 480 ppm males. No females exhibited any sign of hyperactivity. Emaciation occurred in four 240 ppm males, and in three males and in all 480 ppm females. In general clinical findings were more severe and occurred earlier in the 480 ppm as compared to the 240 ppm dose group. Emaciation was observed more frequently in the females than in males. Emaciation and hyperactivity appear to be related to treatment.

6

2. Body Weight

Summary mean body weights are presented in Appendix B. Body weights decreased with increasing doses, and the differences between mean body weights of treated and control groups increased over the 28 day study period. By day 28, body weights of the 480 ppm and 240 ppm groups were lower than the controls. The males were 42.1% and 34.9% lower, respectively as compared to the control, while the females were 50.2% and 20.8% lower, respectively as compared to the control. By day 28, the 120 ppm female body weights were 15.6% lower than controls. Body weights of other treated groups were comparable with the controls.

3. Food Consumption

Mean weekly food consumption data are presented in Appendix C. The food consumption values of both sexes were lower than their respective controls, at all intervals in the 480 ppm dose group and starting from day 14 in the 240 ppm dose group. By day 28, the food consumption values of the 240 ppm group were 50% (males) and 36.5% (females) lower, and in the 480 ppm group they were 65.6% (males) and 59.7% (females) lower than their respective controls. By day 28, the food consumption value in the 120 ppm female dose group was 26.5% lower than the control. Reduced food consumptions in both sexes of the 240 ppm and 480 ppm rats, and in the 120 ppm female rats are related to treatment.

4. Testis and Epididymis Weights

Mean absolute and relative testis and epididymis weights are presented in Appendix D. The relative testis weights of the control were comparable with 30, 60, 120 ppm groups. The relative testis weights of the 240 ppm (two rats) and 480 ppm (one rat) dose groups were markedly higher (44.3% and 50.0%, respectively), than the controls. The relative epididymis weight of the 240 ppm group was 35.0% higher than the controls. Since the relative testis and epididymis weight increases were attributed to the reductions of body weights, they are not considered to be treatment-related effects.

5. Gross pathology

Summary gross necropsy observations are presented in Appendix E. The most notable finding was the thickening of the liver, one in the control, three in the 30, one in the 60, and three in the 120 ppm dose groups. Discoloration of the liver was observed in two 240 ppm male and in one male and one female of the 480 ppm dose groups. Undescended testes were observed in three 480 ppm and one 240 ppm males.

6. Histopathology

Summary of microscopic findings are presented in Appendix F. The two grossly abnormal 240 and 480 ppm livers were microscopically determined to be hepatocellular hypertrophy. One of the two 240 ppm livers also exhibited hepatocellular vacuolation and one of the two 480 ppm rats also exhibited multifocal necrosis of the liver. It appears that the liver is the target organ of "sulfluramid 20/80". Although it was stated (p. 14 of the study report), that grossly abnormal livers in the 120 ppm and 240 ppm dose groups were microscopically evaluated, the investigators failed to evaluate grossly abnormal livers of the 120 ppm and other lower dose groups (see attached Appendix E).

G. SUMMARY

Six groups of five males and five females of Sprague-Dawley rats were fed with 0, 30, 60, 120, 240, and 480 ppm sulfluramid 20/80 in the diet for a period of 28 days.

Sulfluramid 20/80 related effects included reduced survival, lower body weights and food consumption, and increased clinical signs of toxicity when compared to controls. These effects were observed in the 240 ppm and 480 ppm dose groups.

Body weights were reduced with increasing doses, and the difference between mean body weights of treated and control groups increased over the 28 day study period. Reductions in body weights noted in the 240 ppm and in the 480 ppm dose groups of both sexes were judged to be related to treatment.

Reduced food consumptions in both sexes of the 240 ppm and 480 ppm rats, and in the 120 ppm female rats are related to treatment.

A notable gross finding was the thickening and discoloration of the liver. Increased incidences were noted in the treated groups as compared to the controls. The two grossly abnormal 240 and 480 ppm livers observed during necropsy were determined to be a hepatocellular hypertrophy. It appears that the liver is the target organ for "sulfluramid 20/80".

Based on mortality, body weight, food consumption, and clinical signs, treatment-related effects were observed in the 240 and 480 ppm rats of both sexes as well as in the 120 ppm females. The data also show that females appeared to be more sensitive than males.

The definitive NOEL and LOEL can not be determined because not all livers were microscopically evaluated and the number of animals used were too small. Based on body weight and food consumption data, the LOEL is determined to be 240 ppm for the males and 120 ppm for the females, and the NOEL is 120 ppm for the males and 60 ppm for the females.

There is some concern that livers of the intermediate dose groups (30, 60, and 120 ppm) were not histopathologically evaluated. Furthermore, the test article analysis showed that it contained a 31.7% linear and a 62.6% branched Sulfluramid isomers. This mixture deviated somewhat from the intended ≈20% linear and ≈80% branched isomers mixture indicated in the study report.

The investigators should clarify why some livers of the intermediate dose groups were not evaluated and also why the linear and branched Sulfluramid isomer mixture based on the analysis deviated from the intended isomer ratio.

Sulfluramid

Page____ is not included in this copy.

Pages 9 through 13 are not included.

The material not included contains the following type of information:

____ Identity of product inert ingredients.

____ Identity of product impurities.

____ Description of the product manufacturing process.

____ Description of quality control procedures.

____ Identity of the source of product ingredients.

____ Sales or other commercial/financial information.

____ A draft product label.

____ The product confidential statement of formula.

____ Information about a pending registration action.

X FIFRA registration data.

____ The document is a duplicate of page(s) _____.

____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.