1-24-80

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY

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SUBJECT: EPA Reg.#241-243; Prowl; Oral Teratology Study in Rats. CASWELL#454BB; Accession#241595

FROM: William Dykstra
Toxicology Branch (TS-769) WHD 1/25/80

TO: Robert Taylor
Product Manager#21

THRU: M. Adrian Gross, Chief William M. Dykstra Jr. M. Adrian Gross
Toxicology Branch (TS-769)

Recommendations:

1. The rat teratology study is acceptable as core-guideline data. Ac 92,553 is not teratogenic or fetotoxic at doses up to 500 mg/kg/day.

Review:

1. Oral Teratology Study in Rats; AC 92,553, Final Report (Hazelton Project No. 362-155, 8/17/79)

Test Material

Seventy male rats and one hundred forty female rats of the Sprague-Dawley CD strain were used in the experiment. The body weights of the males ranged from 250 to 300 grams and the body weights of the females ranged from 180 to 200 grams.

The rats were uniquely identified by ear tags, and were individually housed in wire-mesh cages in an air-conditioned room with a 12 hour light-dark cycle. Water and Purina Lab Chow were available ad libitum.

Following a two-week acclimation period in the laboratory and examination for health by a staff veterinarian, the rats were mated by placing one male and two females in a breeding cage until a sufficient number of females were impregnated (no longer than three weeks).

A vaginal examination of each female was performed daily with a microscopic examination of a slide prepared from a small drop of normal saline delivered into and then recovered from the vagina with a pipette. The slide was examined for the presence and viability of sperm. The day of observation of sperm or vaginial plug was designated as Day 0 of gestation, and the females were then assigned consecutively to the experimental groups until a sufficient number of mated females were present in each group.

Group	Number of Mated females	Number of pregnancies required	Dosage levels
1 (control)	33	25	corn oil
2 (low)	34	25	125 mg/kg
3 (mid)	33[a]	25	250 mg/kg
4 (high)	33	25	500 mg/kg

[a] Animal number 75944, Group 3 female, was discarded from the study on May 16, 1979 because she received Group 4 compound. Statistics are based on 32 mated females in Group 3.

Appropriate amounts of AC 92,533 were mixed with corn oil to form a suspension. A sufficient volume of each dosing solution for the entire study was prepared initially and reserved samples (10 mls) were forwarded to the sponsor for analysis. The sppropriate volume of the test material or corn oil alone was administered to the females by oral intubation daily (at approimately the same time each day) from Day 6 through Day 15 of the gestation period. The daily dosage of the test material was based on the individual rat's body weight on the first day of administration.

All of the animals were observed daily for appearance, behavior, mortality and moribundity. Individual body weights were recorded on Days 0, 6, 9, 12, 15 and 20 of gestation, and food consumption was recorded on Days 6, 9, 12, 15, and 20 of gestation. On day 20 of gestation, all surviving rats were sacrificed by carbon dioxide asphyxiation, and a gross necropsy was performed. All observed fetuses were removed by cesarean section and the number of corpora lutea, implantation sites, resorptions, live fetuses and dead fetuses in each uterine horn were recorded.

The fetuses were individually removed from the placenta, identified, weighed, externally examined, sexed, and the crown-rump distance was measured and recorded.

The males were anesthetized and sacrificed without necropsy upon completion of mating.

One-third of the fetuses from each litter were fixed in Bouin's solution, sectioned by Wilson's freehand razor technique, and sealed in plastic. Whole body transverse sections of the head, thoracic and abdominal regions were taken and examined for visceral abnormalities with the aid of magnification.

The remaining two-thirds of the fetuses from each litter were examined internally, eviscerated, and placed in 95% ethanol. After proper fixation and dehydration, the skeletons were stained in a potassium hydroxide-alizarin red solution and finally cleared in a solution of glycerol-ethyl alcohol and benzyl alcohol, and stored in glycerol ethanol (50/50). Each skeleton was examined with the aid of magnification on a light box for ossification, bone alignment and anomalies.

The following were the procedures for the preservation of the various tissues: (1) the ovaries and uterus of each rat were preserved in 10% neutral buffered formalin; (2) the fetuses examined by Wilson's technique were preserved in Bouin's solution and sealed in plastic after sectioning; (3) the fetuses stained for skeletal examination were preserved in plastic in the glycerol and ethanol (50/50) solution with several crystals of thymol to retard bacterial growth.

Statistical Analyses of the data were performed by various methods. All analyses were evaluated at $p < 0.05$.

Results:

Cloudy eyes, sores on the tail, and scales on the tail were the most frequently reported clinical observations in the dams. These observations occurred with a comparable frequency in all groups and are considered incidental.

Urine stains occurred more frequently in the mid- and high-dose rats on days 6-15 than in the low-dose and control animals.

Analyses of mean maternal body weight changes revealed no statistically significant differences between the control and test groups calculated for days 0-6, 6-15, 15-20, 0-20 of gestation. Analyses of maternal food consumption revealed a statistically significantly higher mean value for the high-dose group when compared to the control group on day 20 of gestation. No other statistically significant differences between the treated and control groups were noted at any of the other days analyzed.

At sacrifice (day 20); the uterus was distended with clear fluid in two control rats, two low-dose rats, and one mid-dose rat. All five of these females were found not pregnant on day 20 of gestation. One low-dose rat had a tissue mass on its liver, and one high-dose rat had a nodule in the inguinal area. Two low-dose rats, three mid-dose rats, and eight high-dose rats had a yellow tinge to their body fat. Besides the greater incidence of yellow body fat in the high-dose group, no treatment-related tendencies were noted.

Pregnancy rates were 85%, 88% and 91% for groups 2, 3, and 4, respectively, and 88% for the control group.

There was a significantly higher mean number of corpora lutea in the high-dose group when compared to the control group; however, there were no significant differences noted between the treated and control groups with respect to the mean number of implantations, and implantation efficiencies.

There were no statistically significant differences between the treated and control groups for the incidence of resorption, fetal death or fetal viability. The mean fetal lengths and weights were not significantly different when the treated groups were compared to the control group.

The visceral examination revealed hydronephrosis in one mid-dose fetus. No other visceral anomalies were noted. The skeletal examinations revealed anomalies of the ribs and vertebral column in one high-dose fetus. No other skeletal anomalies were noted. Incidental findings among control and treated groups of logging ossification of the skull, rib cage, vertebral column pelvic girdle, and extremities and angulated ribs were also observed. These findings are considered to be common variants seen in rats at Hazelton Labs.

Conclusion:

AC 92,553 was not teratogenic in rats when given at dosages up to 500 mg/kg during days 6-15 of gestation. The NOEL for fetotoxicity is also 500 mg/kg/day.

Classification: Core-Guideline DATA

TOX/HED:th:RD Initial:CFRICK:1-25-80

C. Frick
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