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3-3-81

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

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OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: EPA Reg.#4581-EUP-32; Hydout™ Aquatic Algicide and Herbicide
CASWELL#421; Accession#244125

FROM: William Dykstra, Toxicologist
Toxicology Branch, HED (TS-769) (WD for LOC 3/3/81

TO: Richard Mountfort (23)
Registration Division (TS-767)

Recommendations:

1) The label signal word and precautionary labeling are incorrect. Based on review of acute toxicity studies submitted previously in request for permanent tolerances (memo of 10/25/78 from W. Dykstra to H. Jacoby), Hydout has the following acute toxicity profile:

Primary eye irritation: Toxicity Category I: Danger
Primary skin irritation: Toxicity Category IV: Caution
Acute oral LD_{50}: Toxicity Category III: Caution
Acute dermal LD_{50}: Toxicity Category III: Caution

On the basis of the results of the primary eye irritation, the label signal word is Danger (rather than Caution as now printed) and appropriate precautionary labeling, e.g. corrosive, causes severe eye damage should be added to the label. The product should be considered a candidate for restricted use.

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2) With respect to the rat teratology study, the response of the registrant's consultant, Dr. Clara Williams, and the revised report by FDRL do not satisfactorily address all of the concerns of the reviewer regarding the adequacy of the study to support the requeste EUP or permanent tolerances. If appears possible from the FDRL report that incomplete closure of the skull may be considered cranioschisis rather than delayed ossification. A rat teratology with postnatal evaluation could address the significance of this cranial finding. In addition, the number of fetues affected compared to the number of fetuses examined are 4.6%, 25%, 14.6% and 9.5% for the control, low, mid, high-dose levels, respectively. These results, though not dose-related, demonstrate that the control and high-dose fetuses did not respond the same as indicated by Dr. Williams. The reviewer concurs with FDRL and Dr. Williams regarding the significance of the 14th rib, the incidence of which is low in the control and Endothall treated groups. In addition, the reviewer concurs with Dr. Williams that interference with implantation at the 2 mg/kg dose level does not seem likely since implantation occurred prior to the administration of Endothall.

   A repeat of the rat teratology study with postnatal evaluation portion is necessary to address the question of the teratogenic or fetotoxic potential of Endothall in the rat and support the EUP.

3) The toxicology studies submitted are acceptable as Core-Minimum Data or Supplementary Data.

Review:

1) Exhibit 190.

   This exhibit consists of:

   a) Letter from Clara Williams - December 31, 1980
   b) Clara Williams' Curriculum Vitae
   c) Letter from Richard A. Parent - January 7, 1980
   d) Historical Control Data on Charles River Rats
   e) FDRL Report 5129 on Endothall
   f) FDRL Report 5129 Protocol

   FDRL report 5129 was previously reviewed in memo of 10/25/78 from W. Dykstra to H. Jacoby. With respect to the rat teratology study, the response of the registrant's consultant, Dr. Clara Williams and the revised FDRL report do not satisfactorily address all of the concerns of the reviewer regarding the adequacy of the study to support the requested EUP or permanent tolerances. It appears possible from the FDRL report that incomplete closure of the skull may be considered cranioschisis rather than delayed ossification. A rat teratology study with postnatal evaluation could address the significance of this cranial finding. In addition, the number of fetuses affected compared to the number of fetuses examined are 4.6%, 25%, 14.6% and 9.5% for the control, low, mid, high-dose levels, respectively. These results, though not doserelated, demonstrate that the control and high-dose fetuses did not respond the same as indicated by Dr. Williams. The reviewer concurs with FDRL and Dr. Williams regarding the significance of the 14th ribs, the incidence of which is low in

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the control and Endothall treated groups. In addition, the reviewer concurs with Dr. Williams that interference with implantation at the 2 mg/kg dose level does not seem likely since implantation occurred prior to the administration of Endothall.

Conclusion:

A repeat of the rat teratology study with a postnatal evaluation portion is necessary to address the question of the teratogenic or fetotoxic potential of Endothall in the rat and to support the EUP.

Classification: Supplementary Data

2) Exhibit 191.

Test for Guinea Pig Sensitization (MB Research Laboratories, Inc., Project No. MB-79-4140; 11/6/79)

Test Material: Hydout Aquatic Algicide and Herbicide

Hartley Albino guinea pigs, approximately 5 weeks old when received were equilibrated for at least one week. Ten apparently healthy guinea pigs were selected for the test. The dorsal area of each animal was clipped free of hair prior to the first treatment of each week. The test material was applied dermally 3 consecutive days/week for three weeks and again on the first day of the series in the fourth week. The initial application and the change dose were 0.05 ml; the remaining nine applications were 0.1 ml. The application sites were rotated so that no one site was dosed twice. Fourteen days after the last application exposure, the animals were challenged in the same manner at a site removed from the sites of application treatments. Dermal reactions were scored at 24 hours after each treatment and challenege by the Draize scoring system. Average redness and edema scored in all animals at 24 hours after each treatment were compared to the average redness and edema scores in all animals 24 hours after the challenge. A material was considered to be sensitizing if a significant increase in scores was noted.

Results:

Average erythema of 10 applications was 1.5 and average edema was 0.9. At challenge, the average erythema was 0.6 and average edema was 0.1.

Conclusion:

Under the conditions of this test, the test material is not a sensitizer.

Classification: Core-Minimum Data

3) Exhibit 192.

Evaluation of Herbicides for Possible Mutagenic Properties; K.J. Andersen, E.G. Leighty and M.T. Takahasi; J. Agr. Food Chem.; Vol. 20, No. 3, 1972 (pages 649-656); Author's Abstract.

One hundred and ten herbicides were evaluated for their ability to induce point mutations in one or more of four difference microbial systems. None of the herbicides appeared to cause point mutations in these microbial system in comparison with known mutagen such as 5-bromouracil or 2-aminopurine. Except for inconclusive evidence relating to four herbicides (endothall not included) withith one test of one system, mutagenic rates of herbicide-treated organisms did not differ significantly from spontaneous rates. In this one test, four herbicides were associated with mutation frequencies slightly in excess of the control. The observed increases were small, and the rates of mutation were lower than spontaneous rates of controls in other tests of the same system. Therefore, it appears that the increases observed with these four herbicides were within the normal range of spontaneous rates.

Classification: Core-Minimum Data

4) Exhibit 193.

24-Month Study of Disodium Endothall (15.8% active) in CD, Mice; Addendum I (gross pathology); Addendum II (protocol) (Cannon Labs#6E-4479; 6/22/79)

A total of 400 mice, approximately four weeks of age, were obtained from the Charles River Laboratories, Inc., Wilmington, Mass. The weight range of experimental animals at week 0 was 23.3 + 0.24 gm for females and 28.4 + 0.34 gm for males. When received, each animal was ear-tagged and was identifiable by a unique number. If this tag was lost, the animal was identified by a unique combination of ear punches. Animals were then housed by sex in groups of 5.

Test animals were randomly assigned to one four treatment groups as shown below:

Group	Dose level (ppm)	Number of Animals	
		Males	Females
I	0	50	50
II	300	50	50
III	600	50	50
IV	1200	50	50

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All animals were examined daily for general physical appearance, palpable masses and pharmacotoxic signs. All body weights were recorded at weekly intervals through week 12, and monthly thereafter for the duration of the study. Means per cage were used as the base of measure, resulting in each cage having a representative animal weight expressed in grams.

Food consumption was recorded at weekly intervals through week 13, and monthly thereafter for the duration of the study. Consumption was recorded as the mean value per cage, in grams.

The heart, kidneys, spleen and gonads of each animal surviving to termination were weighed to the nearest 1×10^{-3} gm. Liver weight measurements were overlooked and were not taken. These measurements were also represented as an index obtained by dividing the organ weight by the final body weight.

Gross necropsies were performed on all non-autolyzed or non-carnnibalized animals. Tissues for histopathology were removed from animals surviving to termination, sacrificed in extremis, or found dead, but not autolyzed (a total of 201 animals). Additionally, some tissues were taken from animals found dead but partially autolyzed (a total of 129 animals). No tissues were available from 64 animals (totally autolyzed) and 6 animals (totally cannibalized). After gross pathological examination, all available tissues were preserved in 10% neutral buffered formalin for future microscopic examination.

adrenal
bone marrow (sterum)
brain (3 levels)
duodenum
esophagus
eye and Harderian gland
gall bladde
heart
kidney
liver
lung
lymph nodes
mammary glands
ovary/testes
pituitary
pancreas
prostate/uterus
salivary gland
sciatic nerves
skin
small & large intestines
spiral cord
spleen
stomach
thymus
thyroid/parathyroid
trachea
urinary bladder
gross lesions
Vertibra Tibia Femoral Joint

Also, the nasal cavity, tongue, oral cavity, nasopharynx and middle ear were removed from ten survivors per sex per group. If fewer than ten animals survived, all animals alive at termination in that group had these tissues removed.

Statistical analyses of the data were performed.

Results:

No effect of the test material on survival was apparent in either males or females. At week 70, approximately 20% of the males had died in each group. This pattern continued through week 103, at which time 14% (controls), 12% (300 ppm), 6% (600 ppm) and 26% (1200 ppm) of the males had survived. Females followed a similar pattern through week 79 although the total number of females surviving at termination appeared to be somewhat greater than that of the males; 38% (control), 24% (300 ppm), 34% (600 ppm) and 50% (1200 ppm) of the females had survived.

Males did not appear to exhibit obvious differences in frequencies of pharmacotoxic signs of treatment. The time at which these signs were first noted also appeared to be similar. Likewise, the frequency, time of recognition and duration of pharmacotoxic signs in females did not appear to differ among treatment groups.

Thickened conjunctiva, however, were noted in 34 males but only 6 females, and lacerations were seen in 23 males versus nine in females.

Masses were found in the stomachs of females treated with test material on 15 occasions. The stomachs of control females appeared normal in all cases. Similarly, pancreatic masses were evident in five test material treated females; however only one mass on the pancreas of control females was found. Masses of the lung were seen in ten females receiving 1200 ppm of test material in the diet compared with frequencies of 4, 5 and 7 for females receiving the control, 300 and 600 ppm diets, respectively.

Summing over sex, high dose animals exhibited nearly twice as many internal masses as controls (50 high-dose animals vs 30 control animals).

Significant differences were found in body weights among females at various times throughout the 24-month study. However, these differences were sporadic. A more clear pattern of pattern of differences was seen in the body weights of males. Animals treated with the test material frequently weighed less than controls through week 77. The body weight of controls showed marked fluctuations from week 77 through termination at week 103.

Differences of average food consumption among females did occur; however, no clear dose-related response was obvious. Few differences were seen in food consumption among males.

The heart to body weight ratio in group III (600 ppm) females was lower than compared to group I (0 ppm), II (300 ppm) and IV (1200 ppm). Group IV males exhibited higher gonad weight to body weight ratios and gonad weights than males receiving 0, 300 or 600 ppm of test material in the diet. Males in groups III and IV had lower kidney weights than those in group II and I. No other organ weight differences of statistical significance were observed.

Conclusion:

No conclusions regarding the systemic or oncogenic effects of the test material can be made until the report on the histologic examination of the masses and tissues is reviewed.

Classification: Supplementary Data

5) Exhibit 194.

Activity of T1604 in the Salmonella/Microsomal Assay for Bacterial Mutagenicity (Microbiological Associates; April 29, 1980) T1604 = Endothall

T1604 was tested in the Salmonella/microsomal assay in the strains TA-1535, TA-1537, TA-1538, TA-98 and TA-100 at five dose levels: 5 mg, 1.5 mg, 0.5 mg, 0.15 mg and 0.05 mg/plate in the absence and presence of exogenous metabolic activation. Positive controls were tested.

Results:

No detectable mutagenic activity was found for the test material under the conditions of the assay.

Classification: Core-Minimum Data

6) Exhibit 195.

Activity of T1604 in the in vitro mammalian cell point mutation assay in the absence of exogenous metabolic activation (Microbiological Associates; 6/23/80)

The purpose of ~~this study~~ was to employ the BALB/3T3 clone A31 mouse cell line in the absence of an exogenous metabolic activation system in order to investigate the in vitro mutagenic potential of the test material. Endothall was tested at doses of 10 ug/ml, 3 ug/ml, and 1 ug/ml. Positive controls were tested.

Results:

The negative control and the positive control fulfilled the requirements for determination of a valid test. The criterion established for the surviving fraction of cells treated with the test material was also satisfied. Statistically significant ($P < 0.05$) mutagenic activity was not detectable for Endothall under the conditions employed in the assay.

Classification: Core-Minimum Data

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7) Exhibit 196.

Activity of T1604 in the in vitro mammalian cell point mutation assay in the presence of exogenous metabolic activation (Microbiological Associates; 6/26/80)

The purpose of this study was to employ the BALB/3T3 clone A31 mouse cell line in the presence of an exogenous metabolic activation system in order to investigate in vitro the mutagenic potential of the test material. Endothall was tested at doses of 1 ug/ml, 0.1 ug/ml and 0.01 ug/ml. Positive controls were tested.

Results:

The negative control and the positive control fulfilled the requirements for determination of a valid test. The crierion established for the surviving fraction of cells treated with the test material was also satisfied. Under the conditions employed in the assay, no detectable mutagenic activity was found for Endothall.

Classification: Core-Minimum Data

8) Exhibit 197.

Endothall Range-Finding Study in Non-Pregnant Female Mice (IRDC Report No. 470-004, no signatures, no date, draft)

Sexually mature virgin female Charles River CD-1 mice were used to establish dosage levels of Endothall for a pilot teratology study. Dosage levels of 30, 60 and 100 mg/kg/day were administered orally by gavage at a single daily dose on days 1 through 5 at a constant volume of 5 ml/kg. The control group received the vehicle only, deionized water, on a comparable regimen at a constant volume of 5 ml/kg. Necropsies were performed on all surviving females on study day 6.

Results:

Survival was 100% in the control group and in the 30 mg/kg/day dosage group. There were no remarkable findings in the appearance, behavior or the postmortem examination observations in any females in the 100 mg/kg/day dosage group and four of the five females in the 60 mg/kg/day dosage group which died on study day 2. The remaining female in the 60 mg/kg/day dosage group died on study day 5.

A cause of death could not be determined for any of these mice. There was no mean gain in body weight over the treatment period in the 30 mg/kg/day dosage group.

Conclusion:

Based on the results of this study, dosage levels of 5, 10, 20, 40 and 60 mg/kg/day were selected for the pilot teratology study in mice.

Classification: Supplementary Data

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9) Exhibit 198.

Teratology Study in Mice (IRDC#470-006; 8/12/80)

IRDC Protocol for teratology study in mice. Dosages not specified.

Classification: Supplementary Data

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