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

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM FEB 15 1983

Caswell No. 706A

DATE: January 28, 1983

SUBJECT: Safrotin Insecticide (propetamphos, Sandoz) -- Evaluation of Japanese Two-Generation Study in Rats. In re: Food Additive Petition 2H 5349 (EPA Registration No. 11273-23).

TO: William H. Miller, Product Manager (16)
Registration Division (TS-767C)

PETITIONER: Sandoz Inc., Crop Protection, 480 Camino del Rio South, San Diego, CA.

ACTION REQUESTED: Review of two-generation study in rats (reported in three separate segments), performed in Japan by Sandoz Pharmaceuticals Ltd., Tokyo, as teratogenicity and reproductive tests (Report No. CBK I. 4830/80, May 16, 1980), and submitted by Sandoz Inc., November 4, 1982, as a reproduction test in support of their food additive tolerance petition, 2H 5349. [As stated in the Introduction of this submission, it is noted that this study was conducted in Japan for the purpose of obtaining a Japanese registration, since that country only considers registrations that are supported in part by toxicology data generated in Japan.]

TOXICOLOGY BRANCH EVALUATION (SUMMARY): The study submitted is judged SUPPLEMENTARY DATA, because it does not qualify as an adequate reproduction test (nor as a teratogenicity assay) according to the (FIFRA) Pesticides Assessment Guidelines - Subdivision F, Sect. 83-4: "Reproductive and fertility effects" (1983, NTIS), due to the following deficiencies:

(1) Different groups of animals were used for the three segments, and no group was treated through two generations.

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(2) An LEL for any clinical changes was not determined (HDT = 2 mg/kg, stated to decrease plasma cholinesterase), and no clinically observable effects were found at the highest dose.

(3) No measurements of cholinesterase were made in test animals; neither a NOEL or LEL (with or without clinical effects) was determined.

(4) Identity and concentrations of impurities in the test substance (technical) were not provided; purity of the technical was stated as 92%.

(5) Females of the P_1 generation received test substance for only 2 weeks before and during mating (an 8-week pre-mating treatment period is recommended in both sexes).

(6) F_1 animals were not dosed in any segment of these studies.

(7) F_2 animals were not observed to weaning.

(8) Gross necropsies were not performed on the F_2.

[The second segment, Study B of the submitted study might have been acceptable as a teratogenicity assay with post-natal segment with the demonstration of observable maternal clinical effects, and a NOEL for fetal effects in the absence of maternal clinical effects.]

DATA EVALUATION RECORD (REVIEW) follows.

Irving Mauer, Ph.D., Geneticist
Toxicology Branch
Hazard Evaluation Division (TS-769)

2-9-83

cc: L. Chitlik
W. Dykstra

File (2)

2/14/83
2/15/83

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Caswell No. 706A

SAFROTIN (R) (propetamphos, Sandoz) Date: 1/28/83

(DER) DATA EVALUATION RECORD

CHEMICAL: Propetamphos [[(E)-0-2-isopropoxycarbonyl-1-methyl-vinyl 0-methyl ethylphosphoramidothioate]]

FORMULATION: Technical (92%) ,

CITATION: "Propetamphos: Reproduction study in rats." Report No. CBK I. 4830/80, by T. Nakashima, M. Hamada and K. Matsuda, Sandoz Pharmaceuticals, Ltd., Tokyo, Japan, dated May 16, 1980. Performed in three segments, as three separate tests, as follows:

Test A: "EXAMINATION OF EFFECTS OF PROPETAMPHOS ON ANIMALS BEFORE GESTATION AND AT THE EARLY STAGE OF GESTATION"

Test B: "EXAMINATION OF EFFECTS OF PROPETAMPHOS ON ANIMALS AT THE STAGE OF ORGANOGENESIS"

Test C: "STUDY OF THE EFFECTS OF PROPETAMPHOS ADMINISTERED TO RATS DURING THEIR PERINATAL AND LACTATION PERIODS"

TRADE SECRET CLAIM: CONFIDENTIAL BUSINESS INFORMATION.

REASON FOR REVIEW: To support Food Additive Petition, 2H 5349.

REVIEWED BY: Irving Mauer, Ph.D. (TB/HED)

DATE OF REVIEW: January 28, 1983.

TEST TYPE: Oral teratogenicity/reproduction tests in rats.

MATERIALS AND METHODS: JCL-SD male and female rats (25 or more per group per sex) were given test compound in olive oil by gavage at doses of 0.125, 0.5 and 2.0 mg/kg/day (dosage based upon plasma cholinesterase inhibition by preliminary testing in different groups of animals treated for two weeks at 1 and 3 mg/kg/day) in three separate tests: Test A. Treatment of males for 9 weeks and females for 2 weeks before and during mating and gestation, with observations on reproductive indices, and fetal growth and development; Test B. Treatment of pregnant females only (21-23 per group) from gestation day-7 through 17 with observations on dams, and on fetuses from dams sacrificed on gestation day-21 (2/3 of all litters) as well as on pups from spontaneous delivery

(1/3 of litters) postnatally, including F_1 fertility; Test C. Treatment of pregnant females only (22-23 per group) from gestation day-17 through postpartum day-21 (weaning), with observations on dams, and on pups delivered spontaneously (all litters) through their mating (F_1 reproductive capacity) and gestation (F_2 fetuses).

REPORTED RESULTS: Observations and findings in all three tests were documented in summary tables and figures, as well as by complete tabulations of individual animal data.

TEST A RESULTS: There were no deaths in any group, and no significant changes induced in body weight, food consumption, gross pathology or fertility in P_1 test animals, except for renal stones and (right) hydronephosis in one dam each of the 2.0 mg/kg group sacrificed on gestation day-21. Mean weights of fetal placentae from the 0.5 and 2.0 mg/kg groups sacrificed on day-21 were slightly lower than control (significantly in the 0.5 mg/kg group, but stated to be within normal background values); no differences were found in the number of corpora lutea, implantations, liver fetuses, fetal weights or sex ratio. One case of ectopis visceralis and one runt ("nanosomia") were recorded among fetuses from one dam on 2 mg/kg, while non-dose-related hydronephrosis, ureteric dilatation, auricular dilatation and retinal folding were reported in singular instances among all groups. Skeletal variations (dumbbell and/or asymmetrical vertebral bodies, asymmetrical/rudimentary or unossified fifth sternebrae, 14 ribs) were observed in all groups including control, but a statistically significant increased incidence ($P < 0.01$) was reported only for asymmetrical vertebrae (73, 98 and 98 fetuses in the 0.125, 0.5 and 2.0 groups, respectively, vs 50 in controls, as recorded in Summary Table A-8).* Average numbers and degrees of ossification of selected bones (proximal phalanges, metacarpals, metatarsals, and sacral vertebrae) were similar in all test groups and control.

TEST A DISCUSSION AND CONCLUSIONS: Although significant changes were noted in test groups (only) for fetal placental weights (lower) and incidence of asymmetrical vertebrae (increased), these alterations were judged by the authors to be non-compound-related, because they fell within the normal background values for this strain of rats, which were obtained commercially

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* Although not stated in the text of the reports (under "Results"), Table A-8 also records significantly increased incidences over controls (= 35 fetuses) for dumbbell-shaped vertebrae at 0.125 mg/kg (59 fetuses, $P < 0.01$) as well as at 0.5 mg/kg (66 fetuses, $P < 0.05$), but not at 2.0 mg/kg (51 fetuses).

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from CLEA Japan, Inc. [N.B.: No data were presented]. The authors concluded that propetamphos administered orally at doses less than 2.0 mg/kg [N.B.: The Executive Summary stated: "...up to..."] to male and female rats before mating and during gestation did not affect reproductive ("genital") functions of treated animals nor growth of fetuses.

REVIEWERS's EVALUATION (TEST A): This segment of the submitted study (TEST A) is judged SUPPLEMENTARY as a reproduction study because of the following deficiencies:

1. Although the males were dosed for 9 weeks before and during mating, females were treated for only two weeks during this period (which included the first 7 days of confirmed pregnancy).

2. Treatment of that portion of the F_1 delivered spontaneously (1/3 of all groups) was not continued throughout gestation and weaning, in order to ascertain any effects of test chemical on offspring (F_1) growth, development and reproductive functions, as well as effects on the F_2.

3. Although skeletal variations significantly different from control were explicity stated in the text of the report (asymmetrical vertebral bodies at all doses), or discovered on inspection of tabulated data (dumbbell vertebrae at the two lower doses), the authors discount these as not compound-related (within background values), but no background data are presented or cited in support of this contention.

4. No clinically or pathological effects were found at the HDT in any animal; hence an appropriate NOEL cannot be drawn from the data for any potential hazard (risk assessment).

5. Tabulation of body weights for individual control male rats is missing from report (sheet marked as page 30 is blank).

Test B Results: No deaths occurred, nor were any specific signs or symptoms observed in treated maternal groups; body weights and food consumption of all treated dams were comparable to controls. [N.B.: This statement is accepted from the text of the report, since the relevant tabulation, Table B-2, p. 68, is entirely missing from this copy of the report.] "Yellowish" liver was found in one control dam, while (right) hydronephrosis and a renal stone observed in one 0.5 mg/kg animal.

Gross external and visceral fetal examination (2/3 of all pregnancies sacrificed on gestation day-21) revealed similar incidences of hydronephrosis, ureteric dilatation,

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ventricular dilatation and/or hypertrophy, auricular dilatation, and retinal folds in all groups. One case of hydrocephalus and one case of unilateral ("lateral") microphthalmia were found in the 0.125 and 2.0 mg/kg groups, respectively. The authors stated that no induced dose-related skeletal changes (flat/asymmetrical/dumbbell vertebral bodies, asymmetrical/rudimentary or unossified fifth sternebrae, 14 ribs, etc.) were found among fetuses from treated dams.* Enumeration and degree of ossification of proximal phalanges, metatarsals, metacarpals and sacral vetebrae in treated groups did not differ from those in controls.

Duration of pregnancy, number of implantation sites, litter number and sex ratio were comparable in all groups, and no significant differences from concurrent controls observed in dam body weight or food consumption. Except for one 2.0 mg/kg dam with a gastric ulcer, no other specific findings were recorded in any test (or control) dam.

There were no external abnormalities in live-born pups (1/3 of pregnancies) in any group, and no differences between treated and control groups before or after adjustment in littermates (for assessment of F_1 fertility); nor in survival rate, pre- or post-weaning body weight; righting, grasping, pupillary, and stimulus-response (pain) ("nociceptive") reflexes, or open field tests (rearing, grooming, etc.); nor in separation of ear auricles or eyelids, appearance of pelage, eruption of incisors, testicular descent (26 days, except for one 0.5 mg/kg male), or vaginal opening (7 weeks).

Necropsy findings in 8-week old F_1 animals were unremarkable, except for single instances of the following: Abdominal adherence and small size of testis in the one 0.5 mg/kg group male (see above) with undescended testicle; bladder ("vesical") stones in one sacrificed F_1 female of the 0.5 mg/kg group and in one moribund 2.0 mg/kg female, respectively; hydronephrosis in one and pelvic dilatation in a further two females from the 0.5 mg/kg group. A slight but consistent decrease ($p < 0.01$) in mean hepatic weight was

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* Summary Table B-5, however, records a significant ($p < 0.01$) decrease in dumbbell vertebrae compared to controls (= 89 cases) in fetuses from dams treated with 0.5 (31) and 2.0 (51) mg/kg, as well as an increased incidence ($p < 0.01$) in undulated ribs in the mid-dose group (10 fetuses, vs 1, 4 and 4 cases in controls, 0.125 and 2.0 mg/kg groups, respectively.

also recorded in F_1 males and females from the 2.0 mg/kg test group.**

Neither copulation nor gestation rates of F_1 treated groups were different from controls. Except for slight decreases ($p < 0.05$) in body weight of F_1 dams from 0.5 mg/kg treated P_1's during gestation (recorded at day-13), no other ponderal changes were found, and no differences between treated and control groups in food consumption.

Necropsies of F_1 dams at gestation day-21 and examination of F_2 fetuses revealed no differences between test and control groups in the number of corpora lutea, implantations sites, live fetuses, or resorptions. One control F_1 dam had (left) hydronephrosis, and another, an enlarged spleen, while single instances of the following pathological changes were noted in F_1 test groups: Renal "sand" (0.125 mg/kg); bladder-wall "thickening," (right) hydronephosis (0.5 mg/kg). Umbilical ("funicular") hernia was found in one F_2 fetus (0.125 mg/kg group).

Test B Discussion/Conclusions: The authors conclude that rat fetuses exposed to propetamphos at doses less than 2.0 mg/kg [N.B.: The Executive Summary states: "...up to 2.0 mg/kg."] during organogenesis was without apparent clinical effect on mothers, and did not induce any clinical effects or pathological changes in growth, development, or fertility of F_1 animals or on F_2 fetuses.

REVIEWERS EVALUATION (Test B):

This segment of the submission appears to be a teratogenicity study with post-natal observations, but does not qualify as a comprehensive teratology assay, and is judged SUPPLEMENTARY, because of the following deficiencies:

(1) No observable maternal clinical effects were noted in dams treated at the HDT.

(2) There apparently is no LEL for any frank fetal effect, either in the presence or absence of maternal effects.

Test C Results:

No abnormal findings attributible to propetamphos were observed in any of the experimental groups.

** Summary Tables B-15 (male) and B-16 (females) also record slight but significant non-dose related absolute weight changes in treatment-derived F_1's for heart, lung, kidneys, testes, and spleens.

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None of the P1 animals died, and no differences recorded, between control and test groups found in body weight gain and food consumption either during pregnancy or during lactation. Maternal delivery data (rate, duration of gestation, numbers of implantations or pups per litter, sex ratio) were similar in all groups.

There were more stillborn pups and post-natal deaths (to day 4) in the control group (12 and 9, respectively) than in any test group (1 and 1 at 0.125; 3 and 2 at 0.5; and 4 and 6 at 2.0 mg/kg, respectively), but no significant differences in body weight or development (with respect to cardinal signs and body weight) among survivors (post-natal day 4 to weaning), nor in sex differentiation (testicular descent; vaginal opening) or nociceptive reflexes. [N.B.: Table C-9, however, does show small but significant increases in food consumption ($p < 0.01$) among 2.0 mg/kg males at 4 weeks, and among females at 7 weeks postpartum, but falling to control levels thereafter.]

Post-mortem examinations of F_1 animals sacrificed at 10 weeks of age revealed isolated pathological changes, as follows: one enlarged kidney in a female of the 0.5 mg/kg group; hydronephrosis in one female and in three males at 2.0 mg/kg. Slightly decreased weights of livers ($p < 0.01$), but of no other organ (lungs, kidneys, heart, spleen, testes, ovaries) were found in the 2.0 mg/kg group only.

No apparent differences in copulatory and reproductive abilities were discerned between any test group and the control of the F_1, nor in gestation weight gain or food consumption. Post-mortem findings in F_1 animals and their offspring (F_2) revealed no significant induced changes in the numbers of corpora lutea, implantation sites, surviving (live) fetuses, or resorptions. Isolated instances of the following were recorded: Hydronephrosis in one F_1 animal in each of the 0.125 and 0.5 mg/kg groups; two "small" F_2 fetuses in the 0.125 mg/kg group; adhesive placentae of two fetuses, one each in females of the 0.5 and 2.0 mg/kg groups. No other abnormalities were recorded in F_2 fetuses.

The authors conclude that the administration of propetamphos at 2.0 mg/kg and less during perinatal and lactation periods had no effect on the delivery of dams, their lactation condition, nor on the growth, development or reproductive abilities of their (F_1) pups.

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REVIEWER's EVALUATION (Test C): This study is judged SUPPLEMENTARY DATA for reproduction assays:

(1) The F_1 animals were not dosed.
(2) The F_2 animals were not observed to weaning.
(3) No necropsies were performed on F_2 animals.
(5) No clincial effect (no LEL) were apparently found in the P_1, F_1 or F_2 at any dose level.