FILE COPY

(12)

OPP OFFICIAL RECORD
HEALTH EFFECTS DIVISION
SCIENTIFIC DATA REVIEWS
EPA SERIES 361

Cwt 2,*

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF
PREVENTION, PESTICIDES AND TOXIC
SUBSTANCES

August 8, 1996

MEMORANDUM

SUBJECT: Carcinogenicity Peer Review Meeting on **Propazine**

FROM: Esther Rinde, Ph.D. E.R.
Manager, Carcinogenicity Peer Review
Health Effects Division (H7509c)

TO: Addressees

Attached for your review is a package on **Propazine** prepared by Dr. William Dykstra.

A meeting to consider the carcinogenicity classification of this chemical is scheduled for **Wednesday August 28, 1996,** at **10:00 am in Room 817, CM2.**

Addressees

S. Irene
W. Burnam
K. Baetcke
K. Dearfield
H. Pettigrew
B. Fisher
L. Brunsman
E. Doyle
Y. Ioannou
M. Copley
W. Dykstra
R. Gardner
R. Hill
Y. Woo
A. Aranda
R. Ross/L. Brennecke

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

OFFICE OF
PREVENTION, PESTICIDES, AND
TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: Peer Review of Propazine - Re-evaluation

P.C. Code: 080808
Tox.Chem No.: 184
40 CFR 180.243

TO: Esther Rinde, Ph.D.
Manager, Peer Review for Carcinogenicity
Science Analysis Branch
Health Effects Division (H7509C)

FROM: William Dykstra, Ph.D., Toxicologist
Review Section I
Toxicology Branch I
Health Effects Division (H7509C)
William Dykstra 8/1/96

THRU: Roger Gardner, Section Head, Toxicologist
Review Section I
Toxicology Branch I
Health Effects Division (H7509C)
Roger Gardner 8/8/96

C. Background Information

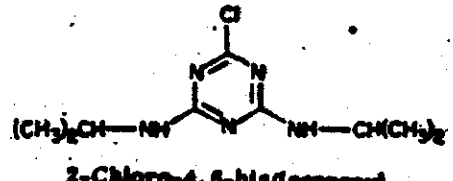
Propazine is 2-chloro-4,6-bis(isopropylamino)-s-triazine. It is a herbicide and controls annual grassy weeds for sorghum. Tolerances are established on sweet sorghum, fodder, forage, and grain at 0.25 ppm.

Propazine has been to the HED Cancer Peer Review Committee (CPRC) twice before. On August 8, 1987, the CPRC first classified propazine as a Group C carcinogen with a Q^*1 of 0.17 $(mg/kg/day)^{-1}$ based on the statistically significant increase in malignant mammary gland tumors in female Sprague-Dawley rats at the 1000 ppm dose level (highest dose tested).

The second CPRC, based on re-evaluations (by the same pathologist) of 3 control and 12 high dose slides in which most the carcinomas in the treated group were rediagnosed as adenomas, classified propazine as a Group C carcinogen without quantification. It was noted that the registration of propazine had been withdrawn by Ciba-Geigy. In light of the discrepancies in the tumor counts, a complete independent re-reading of all the slides would be required, if the reregistration of propazine was ever to be reconsidered.

To comply with the second peer review, the Griffin Corporation completed an independent re-review of all mammary gland slides of female rats in the 2-year rat feeding study, with a draft report dated August 12, 1994. Griffin Corporation then requested a Pathology Working Group (PWG) Peer Review of the proliferative lesions of the mammary glands of female rats be conducted by Experimental Pathology Laboratories, Inc. (EPL) to resolve differences in diagnosis between the original study pathologist and the reviewing pathologist. The PWG was conducted November 30, 1994, and the final report dated January 20, 1995. The results of the PWG are presented in this memo.

STRUCTURE



2-Chloro-4,6-bis(isopropyl-amino)-s-triazine

Propazine

D. Evaluation of Carcinogenicity Data

1. Reference: D. Clifford Jessup; Two Year Oral Chronic Toxicity Study in Rats (April 18, 1981); IRDC #382-007; MRID NO.: 00041408

a. Experimental Design

Randomized groups of 60/sex/dose Sprague-Dawley rats were fed dietary levels of 0, 3, 100, and 1000 ppm (**0.15, 5.0, or 50 mg/kg/day**) for 2 years. An additional 10/sex were added to the control and high dose groups for interim sacrifice at 12 months (5/sex) and a 4 week "recovery period" for 5/sex control and high dose animals. Hematology, clinical chemistry and urinalyses were conducted on 10/sex from control and high dose groups at 3, 6, 12, 18, and 24 months. All animals were necropsied, organ weights were taken at 12 and 24 months and 65/sex from control and high dose were examined microscopically. Mammary gland tissue from all male and female rats in all dose levels was examined microscopically.

LOEL

The NOEL is 100 ppm (5 mg/kg/day). The LEL is 1000 ppm (50 mg/kg/day) and the effect is decreased body weight.

Mammary gland tumors (adenocarcinomas and adenomas) were increased above controls in 3 and 1000 ppm females and were considered compound related. Other tumor types were comparable between control and treated high dose rats of both sexes.

b. Discussion of Tumor Data

1. Mortality Analyses

The statistical evaluation of mortality indicated a significant increasing trend with increasing doses of Propazine in female rats. The statistical evaluation of mortality was based upon the Thomas, Breslow and Gart computer program.

2. Discussion of the Tumor Data

Female rats had significant increasing trends, and significant differences in the pair-wise comparisons of the 1000 ppm dose group with the controls, for mammary gland adenomas and adenomas, fibroadenomas and/or adenocarcinomas combined, all at $p < 0.01$. There was also a significant increasing trend, and significant differences in the pair-wise comparisons of the 3 and 1000 ppm dose groups with the controls, for mammary gland adenocarcinomas, all at $p < 0.05$. In comparison to IRDC historical control tumor data, the incidence of adenomas (17%) and adenocarcinomas (24%) at the high dose of 1000 ppm in the propazine study exceeded the mean incidence for these tumor types (9.5% for adenomas and 11.8% for adenocarcinomas) in the IRDC historical control data, but did not exceed the range for adenomas (2% - 22%) and adenocarcinomas (1% - 29%)

The statistical analyses of the female rats were based upon Peto's Prevalence Test since there was a statistically significant positive trend for mortality with increasing doses of Propazine in female rats.

Table 1. Propazine - Sprague-Dawley Rat Study

Female Mortality Rates[+] and Cox or Generalized K/W Test Results

no increase in male mortality

Dose (ppm)	Weeks 1-26	27-52	53[i]	53-78	79-105[f]	Total
0	1/64[a]	2/62[b]	4/60	8/58	12/50	23/59 (39)**
3	0/60	2/60	0/58	3/58	18/55	23/60 (38)
100	1/60	0/59	0/59	1/59	14/58	16/60 (27)
1000	3/63[c]	0/60	5/60	6/55	24/49	33/58 (57)

[+]Number of animals that died during interval/Number of animals alive at the beginning of the interval.

[i]Interim sacrifice at week 53.

[f]Final sacrifice at week 105.

[a]One accidental death at week 13, dose 0 ppm.

[b]One accidental death at week 52, dose 0 ppm.

[c]Two accidental deaths at week 13, dose 1000 ppm.

( )Percent.

Note: Time intervals were selected for display purposes only.

Significance of trend denoted at control.

Significance of pair-wise comparison with control denoted at dose level.

If *, then $p < 0.05$. If **, then $p < 0.01$.

Table 2. Propazine - Sprague-Dawley Rat Study

Female Mammary Gland Tumor Rates[+] and
Peto's Prevalence Test Results (p values)

	Dose (ppm)			
	0	3	100	1000
Adenomas (%)	1/52 (2)	4/55 (7)	4/58 (7)	9[a]/52 (17)
p =	0.001**	0.127	0.124	0.004**
Fibro-adenomas (%)	20/53 (38)	24/55 (44)	26[b]/59 (44)	24/54 (44)
p =	0.218	0.391	0.347	0.106
Adeno-carcinomas (%)	5/57 (9)	13[c]/58 (22)	8/59 (14)	13/55 (24)
p =	0.047*	0.025*	0.222	0.014*
Combined (%)	23[d]/57 (40)	31[e]/58 (53)	31[f]/59 (53)	37[g]/55 (67)
p =	0.005**	0.124	0.213	0.001**

[+]Number of tumor bearing animals/Number of animals examined, excluding those that died before observation of the first tumor.

[a]First adenoma observed at week 77, dose 1000 ppm.
[b]First fibroadenoma observed at week 71, dose 100 ppm.
[c]First adenocarcinoma observed at week 50, dose 3 ppm.

[d]Three animals in the 0 ppm dose group had multiple tumors.
[e]Ten animals in the 3 ppm dose group had multiple tumors.
[f]Seven animals in the 100 ppm dose group had multiple tumors.
[g]Nine animals in the 1000 ppm dose group had multiple tumors.

Note: Significance of trend denoted at control.
Significance of pair-wise comparison with control denoted at dose level.
If *, then $p < 0.05$. If **, then $p < 0.01$.

A	1	4	6	4	9
AC	5	13		8	13
combined	6	17		11	22

IRDC Historical Controls (1976-1979)

Microscopic Diagnosis	Mean	Range
Adenoma	9.5%	2% - 22%
Fibroadenoma	38.6%	19% - 60%
Adenocarcinoma	11.8%	1% - 29%
Mammary Tumors Combined	50.3%	33% - 68%

c. Nonneoplastic Lesions: There were no nonneoplastic lesions associated with the mammary gland tumors in the propazine rat study.

d. Adequacy of Dosing for Assessment of Carcinogenic Potential

The increasing trend for mortality and the decreased body weight and weight gain in both sexes (> 10%) at the high dose is considered toxicologically significant and evidence that adequate dose levels were used to assess carcinogenicity.

2. Reference: Jessup, D.C. (1980); 2-Year Carcinogenicity Study in Mice; IRDC # 382-004; April 24, 1980; MRID No. 00044335

a. Experimental Design

Randomized groups of 60/sex/dose CD-1 mice were fed in the diet at doses of 0, 3, 1000, or 3000 ppm **(0.45, 150, or 450 mg/kg/day)** for two years with technical propazine.

There were no compound-related effects on mortality, clinical signs, body weight, food consumption or gross pathology. Hematology, urinalysis, clinical chemistries, and organ weights were not determined. At 3000 ppm, an increased incidence of myocardial degeneration was observed in the female mice (17/59 vs 4/60 in controls) and an increased incidence of hemosiderin-laden

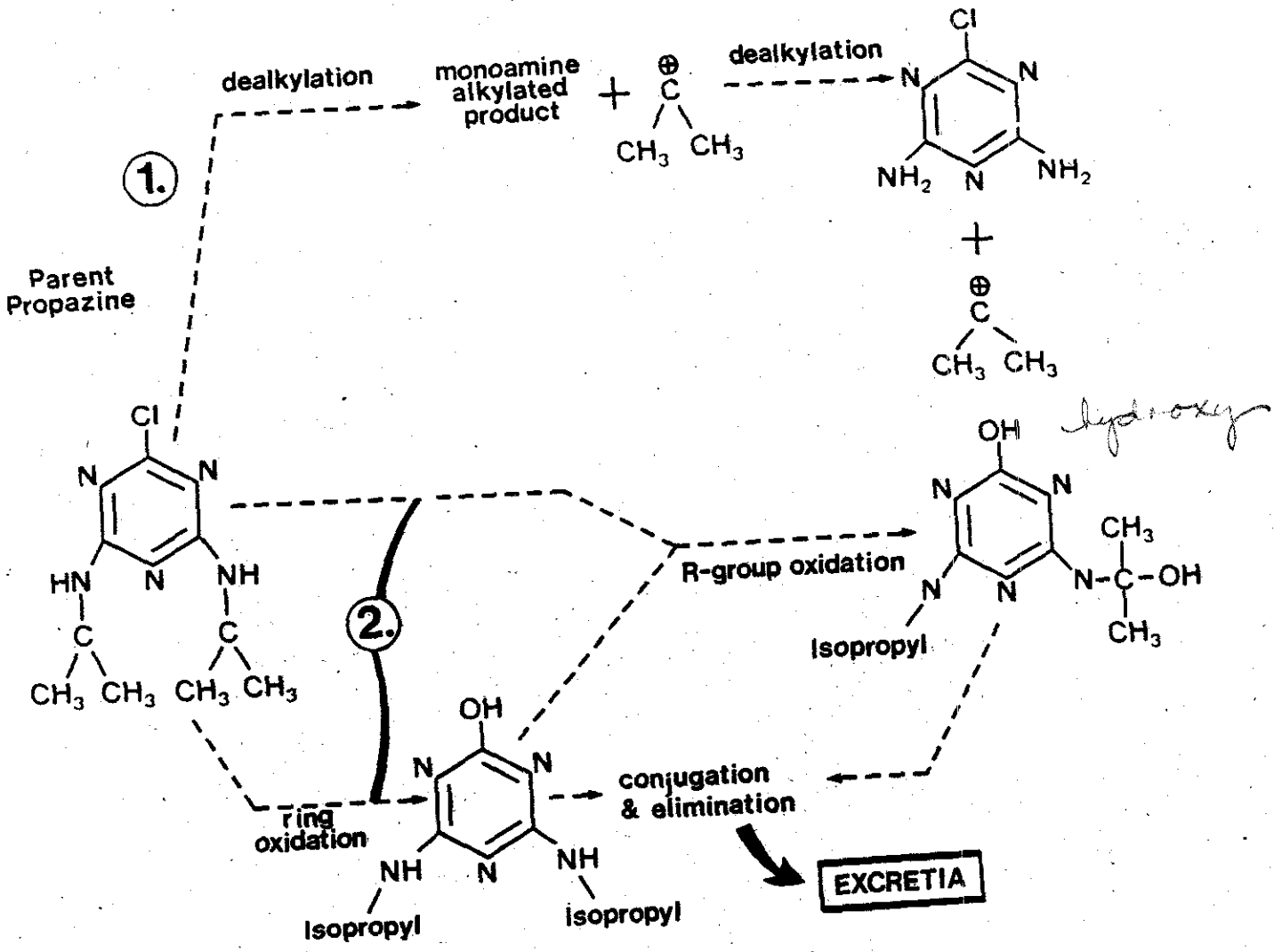
macrophages was observed in the livers of male mice (15/59 vs 3/60 in controls). At the doses tested, there was not a treatment-related increase in tumor incidence. **The NOEL is 1000 ppm (150 mg/kg/day) and the LEL is 3000 ppm (450 mg/kg/day) based on myocardial degeneration in females and hemosiderin-laden macrophages in the livers of males.**

E. Additional Toxicology Data on Propazine

There is a new metabolism study at contractor

1. Metabolism of Propazine

There is no acceptable metabolism study. However, based on open literature information, the general metabolic pathway of propazine in the rat is shown below:



2. Mutagenicity

need dose levels

a. Gene Mutation

Propazine was tested in V79 Chinese hamster cells with microsomal activation and without microsomal activation. Propazine produced a dose-related positive mutagenic response without metabolic activation and a weak (nondose-related) positive response with activation.

b. Nucleus Anomaly Assay

Groups of six male and six female Chinese hamsters were orally administered propazine at dosages of 0, 1250, 2500, and 5000 mg/kg on 2 consecutive days. The cells displaying anomalies of nuclei in treated cells did not differ significantly from the negative controls. Propazine was not considered mutagenic in this assay.

MRID #'s

c. DNA Damage and Repair

Assays for DNA damage in rat hepatocytes were performed with concentrations of 0, 0.5, 2.5, 12.5 and 62.5 ug/mL. The mean number of silver grains per nucleus in the vehicle control and treated cells (at any dose level) was not markedly different. Propazine was not mutagenic for DNA damage and repair under the conditions of this assay.

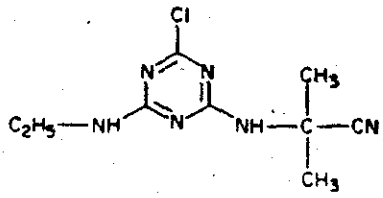
3. Developmental Toxicity

Propazine (25 female Sprague-Dawley rats/dose at doses of 0, 10, 100, or 500 mg/kg/day) was not teratogenic up to 500 mg/kg/day (HDT) [MRID No. 150242]. Maternal toxicity was observed in the mid and high-dose females as decreased food consumption and decreased body weight. Additionally, high dose females exhibited periods of salivation (clear) during gavage. The NOEL for maternal toxicity is 10 mg/kg/day (low dose). Developmental toxicity was observed at the high-dose as increased 14th ribs and incomplete ossification of skeletal structures and decreased fetal body weight. At the mid dose, delayed ossification of the interparietals was observed. The NOEL for developmental toxicity is 10 mg/kg/day.

A developmental toxicity study in rabbits is required.

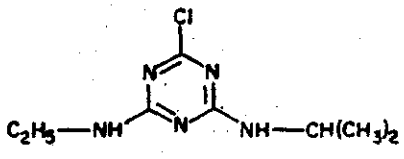
Ten male and 20 female Sprague-Dawley rats/dose were continuously administered dosage levels of 0, 3, 100, or 1000 ppm in the diet for three generations. There were no compound-related effects in male or female fertility, gestation length, pup viability, and survival. Male and female pup weights were significantly reduced at day 21 of lactation at 1000 ppm (HDT) in the F_{1b}, F_{2a}, F_{2b}, F_{3a}, and F_{3b} litters (reproductive NOEL = 100 ppm). Propazine produced systemic toxicity in both males and females including depression in body weights and food consumption and absolute or relative organ weights in all parental animals at the HDT. The systemic NOEL is also 100 ppm.

4. Structure-Activity Relationships



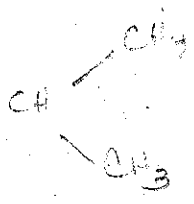
2-Chloro-4-ethylamino-6-(1-cyano-1-methylethylamino)-s-triazine

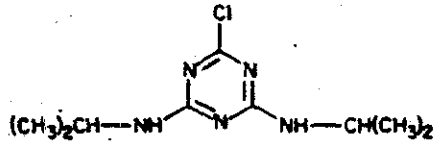
Cyanazine



2-Chloro-4-ethylamino-6-isopropylamino-s-triazine

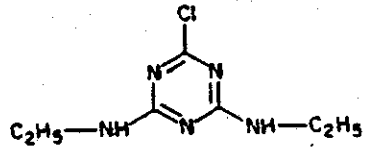
Atrazine





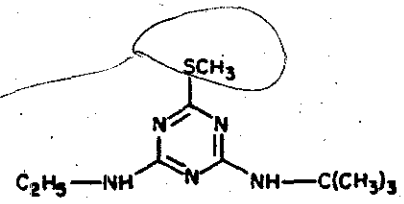
2-Chloro-4,6-bis(isopropylamino)-s-triazine

Propazine



2-Chloro-4,6-bis(ethylamino)-s-triazine

Simazine



2-Methylthio-4-ethylamino-6-tert-butylamino-s-triazine

Terbutryn

Cyanazine was negative in CD mice up to 1000 ppm, but produced a statistically significant increase in malignant mammary gland tumors (adenocarcinoma, carcinosarcoma) in female Sprague-Dawley rats at 25 and 50 ppm. The increased incidence had a significant trend and pairwise comparison and was outside the range of historical controls. Cyanazine was genotoxic in the mouse lymphoma gene mutation assay and unscheduled DNA synthesis in rat hepatocytes. Cyanazine was classified as Group C carcinogen with quantification of human cancer risk using a low-dose extrapolation model (Q^*1).

Simazine was associated with significant increases in carcinomas of the pituitary gland at 1000 ppm (HDT) and carcinomas of the mammary gland at the mid dose of 100 ppm and the high dose of 1000 ppm in Sprague-Dawley rats. Simazine was not associated with an increase in neoplasms in CD-1 mice up to 4000 ppm. There are both positive and negative results in the Ames assay in the published literature. Positive results are reported in the mouse lymphoma, Drosophila sex-linked recessive lethal and cell transformation assays. Simazine was classified as a Group C carcinogen with quantification of human risk using low-dose extrapolation model (Q^*1).

Atrazine was associated with a significant increase in mammary gland fibroadenomas at 1000 ppm, in mammary gland adenocarcinomas at 70, 500, and 1000 ppm and in total mammary gland tumor-bearing animals at 1000 ppm in female Sprague-Dawley rats. Atrazine was not carcinogenic when tested in CD-1 mice. Atrazine was negative for genotoxicity in three acceptable assays, although there are some positive results in the published literature including mouse bone marrow aberrations and a mouse dominant lethal assay. Atrazine was classified as a Group C carcinogen with quantification of human risk using low-dose extrapolation model (Q^*1).

Terbutryn induced a significant increase in combined mammary gland adenomas and carcinomas and combined hepatocellular adenomas and carcinomas in female Sprague-Dawley rats. In males, terbutryn induced an increase in combined thyroid follicular cell adenomas and carcinomas and in testicular interstitial cell adenomas. Terbutryn is negative for carcinogenicity in CD-1 mice and is negative in the Ames assay, chromosomal aberrations in vivo in hamsters, and the micronucleus assay. Terbutryn has been classified as a Group C carcinogen

F. Weight-of Evidence Consideration

The Committee should consider the following facts regarding the toxicology data on propazine in a weight-of-the-evidence determination on carcinogenic potential.

1. The statistical evaluation of mortality indicated a significant increasing trend with increasing doses of Propazine in female rats.

2. Female rats had significant increasing trends, and significant differences in the pair-wise comparisons of the 1000 ppm dose group with the controls, for mammary gland adenomas and adenomas, fibroadenomas and/or adenocarcinomas combined, all at $p < 0.01$. There was also a significant increasing trend, and significant differences in the pair-wise comparisons of the 3 and 1000 ppm dose groups with the controls, for mammary gland adenocarcinomas, all at $p < 0.05$. In comparison to IRDC historical control tumor data, the incidence of adenomas (17%) and adenocarcinomas (24%) at the high dose of 1000 ppm in the propazine study exceeded the mean incidence for these tumor types (9.5% for adenomas and 11.8% for adenocarcinomas) in the IRDC historical control data, but did not exceed the range for adenomas (2% - 22%) and adenocarcinomas (1% - 29%)

need overall conclusion

3. Propazine was tested in V79 Chinese hamster cells with microsomal activation and without microsomal activation. Propazine produced a dose-related positive mutagenic response without metabolic activation and a weak (nondose-related) positive response with activation.

4. Propazine is structurally related to other s-triazines which produce mammary gland tumors and includes terbutryn, atrazine, cyanazine, and simazine.

5. Propazine was negative for neoplasms in CD-1 at doses up to 3000 ppm and was negative in the nucleus anomaly assay and the DNA damage and repair assay.

Study adequate but ~~doesn't~~ doses not excessive.

Attachments

13544

057089

Chemical:	***Propazine***
PC Code:	**080808**
HED File Code	**21200 CPRC**
Memo Date:	**08/08/96**
File ID:	**00000000**
Accession Number:	**412-03-0019**

HED Records Reference Center
12/31/2002