325-5-90

CASWELL FILE

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

JAN - 5 1990

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: Propanil (STAM) - Company Response to Registration Standard - Additional Data on Mouse Oncogenicity Study - EPA Registration Nos. 707-108 and 707-181

Caswell No.: 325
Project No.: 0-0261
Record No.: 255518
MRID Nos.: 412592-01, -02

FROM: William Dykstra, Reviewer William Dykstra 12/15/89
Review Section I
Toxicology Branch I - Insecticide, Rodenticide Support
Health Effects Division (H7509C)

TO: Product Manager Team 74
Special Review and Reregistration Division (H7508C)

THRU: Roger Gardner, Acting Section Head
Review Section I Roger Gardner 12-29-89
Toxicology Branch I - Insecticide, Rodenticide Support
Health Effects Division (H7509C)

Requested Action

Review Rohm & Haas response regarding the issue of bilateral retinal degeneration in male mice and the issue of the MTD in the mouse oncogenicity study with propanil.

Conclusion and Recommendation

One of the toxicological issues relating to the 2-year mouse oncogenicity study has been resolved. TB concludes that there was no compound-related bilateral retinal degeneration in male mice.

However, the HDT of 180 ppm in the 2-year mouse study does not appear to represent an adequate assessment of the MTD. The registrant is required to repeat the 2-year mouse oncogenicity study at considerably higher doses which approximate the MTD (500 to 1000 ppm).

Background

In the December 17, 1987 Toxicology Branch (TB) review (attached) of the company response to the Registration Standard, TB concluded that "TB requires that the grade of the bilateral retinal degeneration lesions in historical controls be provided if available. Although the range of incidence of 2 to 15.5 percent for historical controls encompasses the 11 percent incidence of bilateral retinal degeneration in group 6 male mice, the grade of the lesion of moderately severe in group 6 male mice has not been addressed by the registrant. TB still considers the bilateral retinal degeneration in group 6 male mice to be possibly compound-related. The registrant is invited to submit additional information and/or rationale that may assist in reaching a decision as to whether or not this lesion was induced by the test material."

"TB considers the bilateral retinal degeneration in group 6 female mice not to be compound-related." [End of quotation.]

Additionally, TB concluded regarding the MTD that "The HDT in the 2-year mouse oncogenicity study may not have been the MTD. The HDT was 180 ppm and was based on the results of a 90-day range-finding study in mice. In the range-finding study, severe toxic effects were produced at 1600 and 12,800 ppm. The effects observed at 200 ppm were hepatocytic pleomorphism in 3/10 females and multifocal hepatocellular necrosis in 1/10 males. The HDT for the 2-year mouse study was chosen to be 180 ppm, which is approximately eight times lower than the dose level of 1600 ppm, where 'life-threatening' toxic effects were observed. TB considers the 180 ppm level for the 2-year mouse study to be probably too low and to be based only on hepatocytic pleomorphism and multifocal hepatocellular necrosis, which may be insufficient toxic endpoints. The registrant is requested to provided an explanation and/or rationale as to why the HDT in this study (180 ppm) might be considered the MTD." [End of quotation.]

Review

In response to the TB memorandum, the registrant, Rohm & Haas, has submitted two documents:

1. Response to EPA's Comments Regarding Retinal Degeneration - Report Supplement 82RC-068H (MRID No. 412592-01), and

2. Rationale for Selection of the High-Dose and Justification of the Adequacy of the Study (MRID No. 412592-02)

Issue I - Retinal Degeneration

The occurrences of bilateral retinal degeneration in male mice as observed in the propanil chronic mouse study are shown below.

	Group:	Males					
		1	2	3	4	5	6
Slight		0	1	0	0	1	1
Moderate		2	2	5	1	3	2
Moderately severe		0	0	1	0	0	5s+,S+s+
Severe		0	0	0	0	0	0
Total affected		2	3	6	1	4	8
Number examined		64	63	75	75	79	79
Retina: cannot be examined		9	5	7	10	5	8
Adjusted total examined		55	58	68	65	74	71
% Affected		4%	5%	9%	2%	5%	11%

For males, it can be seen that the grade of the lesion (moderately severe) in five group 6 males (180 ppm, 85.4% technical) is essentially greater than the grade of the lesion (moderate) for most of group 5 males (180 ppm, 98% technical), group 3 males (5 ppm, 98% technical), and group 1 and 2 males (controls, 0 ppm). It can be seen that one group 3 male mouse had a moderately severe grade of lesion in comparison to five group 6 males with this same grade. This difference is not sufficient to negate the conclusion that the incidence of five group 6 male mice with moderately severe retinal degeneration may be compound-related.

Rohm & Haas Reply to Issue I

The registrant's submission states that the grades of severity of retinal degeneration in historical controls were not recorded by the various different Hazleton pathologists. Only the incidence was recorded. The gradings of the lesions are very subjective and it may not be appropriate to compare grades by different pathologists.

The registrant also states that although the incidence of moderately severe bilateral retinal degeneration in group 6 (180 ppm ai Stam technical 85.4%) was increased, the overall incidences of bilateral retinal degeneration in groups 3, 4, and 5 (98% Stam technical) were not dose-related. In fact, the overall incidence of this lesion in group 3 (9%) was comparable to group 6 (11%).

Additionally, when statistical analyses by the registrant of other grades of the lesion are taken into account with the moderately severe grade seen in group 6 mice, the results did not indicate any statistically significant treatment-related linear trend or heterogeneity in the severity of the graded responses.

The registrant also calculates a 19,231 margin of safety between the group 6 dose level (180 ppm) and the TMRC.

TB Conclusion Regarding Issue I

Based on consideration of all available information and analyses, TB concludes that the occurrence of bilateral retinal degeneration in group 6 male mice was not compound-related. This issue is considered resolved.

Issue II - MTD

TB concluded that the HDT in the 2-year mouse oncogenicity study may not have been the MTD. The HDT was 180 ppm and was based on the results of a 90-day range-finding study in mice. In the range-finding study, severe toxic effects were produced at 1600 and 12,800 ppm. The effects observed at 200 ppm were hepatocytic pleomorphism in 3/10 females and multifocal hepatocellular necrosis in 1/10 males. The HDT for the 2-year mouse study was chosen to be 180 ppm, which is approximately eight times lower than the dose level of 1600 ppm, where "life-threatening" toxic effects were observed. TB considers the 180 ppm level for the 2-year mouse study to be probably too low and to be based only on hepatocytic pleomorphism and multifocal hepatocellular necrosis, which may be insufficient toxic endpoints. The registrant is requested to provide an explanation and/or rationale as to

why the HDT in this study (180 ppm) might be considered to MTD.

Rohm & Haas Response to Issue II

Dose selection for the 2-year mouse study was based on the results of 3-month and 2-week range-finding studies. In the 3-month study, hepatocytic pleomorphism and minimal hepatocytic multifocal necrosis were observed at 200 ppm. At the next dose level, 1600 ppm, several effects were observed. These included cyanosis, focal hepatocellular necrosis, pigment in Kupffer cells, hepatocytic pleomorphism, increased relative liver weight and mixed function enzyme activity, splenic extramedullary hematopoiesis and hemosiderin formation, and increased spleen weight.

It was concluded that 1600 ppm would exceed the MDT in a 2-year study.

In the 2-week range-finding study, the NOEL was 1250 ppm and the LEL was 6250 ppm. The LEL did not produce cyanosis. Therefore, it was apparent that the mice were significantly more susceptible after 3 months in comparison to 2 weeks of exposure to propanil.

Therefore, for the high-dose for the 2-year study, the 200 ppm LEL from the 3-month study was reduced by 10 percent to 180 ppm.

Although the liver effects observed at 180 ppm were marginal in the 2-year study, the 1600 ppm level would, as already stated, have exceeded the MTD. The true MTD exists most likely somewhere between 180 and 1600 ppm.

The midpoint of this range is 450 ppm and represents only a 40 percent increase above the 180 ppm level tested.

The nearness of the HDT (180 ppm) to the best estimate of the theoretical MTD (450 ppm) indicates that the oncogenic potential was adequately tested.

Additionally, propanil was not genotoxic in several mutagenicity assays.

The registrant also calculated margins of safety for mixer/loader/applicators (1304) and dietary burden (19,231) resulting from propanil exposure.

TB Conclusion Regarding Issue II

In light of the effects observed at 1600 ppm in the 3-month study and the marginal effects observed at 200 ppm, TB concludes that the HDT of 180 ppm was not an accurate representation of the MTD for the 2-year mouse study. The study has to be repeated at considerably higher doses which approximate the MTD (500 to 1000 ppm).

Attachment

CASWELL

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

DEC 17 1987

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: Propanil - EPA Registration Nos. 707-108, 707-181
Registrant's Response to Letter Dated June 24, 1987
Regarding Toxicological Issues for Propanil

Caswell No.: 325
Record No.: 201958/201959
Project No.: 7-0994
MRID Nos.: 402931-01,
-02, -03

FROM: William Dykstra 12/3/87
Toxicology Branch
Hazard Evaluation Division (TS-769C)

TO: Robert J. Taylor, PM 25
Fungicide-Herbicide Branch
Registration Division (TS-767C)

THRU: Edwin R. Budd, Section Head
Review Section II, Toxicology Branch
Hazard Evaluation Division (TS-769C)

Budd 12/7/87

10/17/87

Requested Action

Respond to additional information regarding propanil chronic mouse feeding study and other toxicological issues.

Conclusions and Recommendations

1. Toxicology Branch (TB) again requests that PM Team 25 (RD) provide full reports of the EPA audit of the propanil chronic mouse study as soon as possible.

7

2. TB requires that the grade of the bilateral retinal degeneration lesions in historical controls be provided if available. Although the range of incidence of 2 to 15.5% for historical controls encompasses the 11% incidence of bilateral retinal degeneration in group 6 male mice, the grade of the lesion of moderately severe in group 6 male mice has not been addressed by the registrant. TB still considers the bilateral retinal degeneration in group 6 male mice to be possibly compound-related. The registrant is invited to submit additional information and/or rationale that may assist in reaching a decision as to whether or not this lesion was induced by the test material.

   TB considers the bilateral retinal degeneration in group 6 female mice not to be compound-related.

3. The occurrence of thyroiditis in group 6 females is not considered compound-related. The incidence in group 6 females is 13.4% (9/67) and is within the range of historical controls of 0 to 23.1%.

4. There are two technicals presently registered with the Agency. A 98% technical registered in April 1985 and an 85-88% technical product registered since 1972. Additionally, a 97% purity technical was used in the 1960s.

   The 2-year rat feeding study, the 2-year dog feeding study, the 3-generation rat reproduction study, the rat metabolism study, and the 90-day rat feeding study were all done in the 1960s by the Medical College of Virginia using a 97% purity technical which may be chemically different from both the 98% and 85-88% technicals registered since that time.

   The rat and rabbit teratology studies were performed with the 85.4% purity technical and were performed in 1980. There are no teratology studies with the 98% technical.

   The mutagenicity studies were performed with either the 85-88% technical or the 98% technical.

   The 2-year mouse oncogenicity study was performed with both the 98% technical (doses of 5, 30, and 180 ppm) and the 85.4% technical (dose of 180 ppm).

Essentially, the 85.4% technical contains [redacted] "total other impurities" which are not present in the 98% technical. These impurities are [redacted]

Additionally, the registrant states: "At the time this study was initated in the first quarter of 1980, and at present, technical Stam(R) was being manufactured for domestic use from [redacted]

"It was desired that this study produce data relevant to the toxicological evaluation of both grades of technical product, and it was therefore decided to include the standard three dose groups of a sample representative of the 98% grade plus an additional high dose group using a sample representative of the 86% grade.

"The three main dose groups using 98% technical would provide a thorough evaluation of the active ingredient common to both grades of technical and of the 98% technical itself. The additional high dose group using the 86% grade material would provide a worst case (high dose) side-by-side comparison of toxic potential with the 98% grade, and would therefore serve to determine whether there were any toxicologically significant differences between the two grades of technical product.

"In the event (see item #3 of this supplement), there were no biologically significant differences between the two technical grades [sic]."

TB Response

TB considers the bilateral retinal degeneration in male mice observed at 180 ppm with the 85.4% purity technical to be possibly a compound-related finding. This finding was not observed at 180 ppm with the 98% technical. Therefore, this compound-related finding may not have a NOEL with the 85.4% technical. The observed effect with 85.4% technical may be due to the "total other impurities."

Issue #2

Historical control data on bilateral retinal degeneration and thyroiditis lesions.

1. The occurrences of bilateral retinal degeneration in male mice as observed in the propanil chronic mouse study are shown below.

Group:	Males					
	1	2	3	4	5	6
Slight	0	1	0	0	1	1
Moderate	2	2	5	1	3	2
Moderately severe	0	0	1	0	0	5s+, S+s+
Severe	0	0	0	0	0	0
Total affected	2	3	6	1	4	8
Number examined	64	63	75	75	79	79
Retina: cannot be examined	9	5	7	10	5	8
Adjusted total examined	55	58	68	65	74	71
% Affected	4%	5%	9%	2%	5%	11%

For males, it can be seen that the grade of the lesion (moderately severe) in five group 6 males (180 ppm, 85.4% technical) is essentially greater than the grade of the lesion (moderate) for most of group 5 males (180 ppm, 98% technical), group 3 males (5 ppm, 98% technical), and group 1 and 2 males (controls, 0 ppm). It can be seen that one group 3 male mouse had a moderately severe grade of lesion in comparison to five group 6 males with this same grade. This difference is not sufficient to negate the conclusion that the incidence of five group 6 male mice with moderately severe retinal degeneration may be compound-related.

The historical control data provided by the registrant are shown below.

HISTORICAL CONTROL DATA
Eyes - Retinal Degeneration

	Males	Females
Unilateral		
Terminal	0/59	2/43 (4.6%)
Unscheduled	0/41	0/57
Terminal	1/24 (4.2%)	0/28
Unscheduled	0/26	0/21
Terminal	2/25 (4.0%)	1/30 (3.3%)
Unscheduled	2/32 (6.2%)	1/26 (3.8%)

HISTORICAL CONTROL DATA
Eyes - Retinal Degeneration (cont'd)

	Males	Females
Bilateral		
Terminal	1/25 (4.0%)	0/26
Unscheduled	0/25	0/24
Terminal	1/24 (4.1%)	0/28
Unscheduled	0/26	0/21
Terminal	7/26 (26.9%)	6/30 (20.0%)
Unscheduled	2/32 (6.2%)	4/26 (15.3%)

Although the range of the incidence of bilateral retinal degeneration (2 to 15.5% for combined terminal and unscheduled deaths) in males encompasses the incidence of 11% in group 6 males, the grade of the lesion in the historical control data was not provided. The grade of the bilateral retinal degeneration in historical controls is requested to be submitted. Unilateral retinal degeneration in male mice in the mouse oncogenicity study was unrelated to treatment.

2. In female mice, the occurrence of bilateral retinal degeneration in the propanil chronic mouse study is shown below:

	Females					
Group:	1	2	3	4	5	6
Slight	0	1	0	1	0	0
Moderate	0	1	3	1	1	4
Moderately severe	0	0	4 S+s+	1	0	3
Severe	0	0	0	1	0	0
Total affected	0	2	7 S+,S+s+	4	1	7 S+,S+s+
Number examined	63	66	80	78	79	79
Retina: cannot be examined	6	6	4	4	3	7
Other adjustments*	0	0	0	0	0	1
Adjusted total examined	57	60	76	74	76	71
% Affected	0%	3%	9%	5%	1%	10%

*Animal A87975.

Both the grade (moderate and moderately severe) and incidence (7) of bilateral degeneration between group 3 females (5 ppm, 98% technical) and group 6 females (180 ppm, 85.4% technical) are comparable.

Since the occurrence of bilateral retinal degeneration in groups 1 and 2 (0 ppm) through group 5 (180 ppm, 98% technical) is not dose-related, it is concluded that the occurrence of bilateral retinal degeneration in female mice tested with 98% technical is not compound-related.

Likewise, since group 3 female mice and group 6 female mice show comparable eye lesions, the occurrence of bilateral retinal degeneration in group 6 female mice (180 ppm, 85.4% technical) is also not considered compound-related.

In support of this conclusion, the (combined terminal and unscheduled deaths) historical control incidence range of 0 to 17.8% for female mice with bilateral retinal degeneration encompasses the incidences (1 to 10%) observed in the propanil chronic mouse study. Unilateral retinal degeneration in female mice in the mouse oncogenicity study was unrelated to treatment.

3. The occurrence of thyroiditis in female mice in the propanil chronic mouse study is shown below:

	Females					
Group:	1	2	3	4	5	6
Number Examined	54	54	68	67	68	67
Thyroiditis						
Not affected	52	52	65	61	66	58
Minimal	1	2	2	3	1	8S+,S+s+
Slight	1	0	1	3	0	0
Moderate	0	0	0	0	0	1
Moderately severe	0	0	0	0	1	0
Total Affected	2	2	3	6	2	9S+s+
Percentage Affected	4	4	4	9	3	13

Statistically significant increases in group 6 females (180 ppm, 85.4% purity) for minimal thyroiditis and total number of animals affected with thyroiditis were observed.

The historical control incidence of inflammation of the thyroid gland in moribund, found dead, and terminally sacrificed CD-1 mice is shown below:

Study	Diagnosis	Incidence/Number Examined Male	Female
	78-Week Dietary Studies		
A	Nonsuppurative thyroiditis	0/99	1/99
B	Mononuclear inflammation	0/70	1/70
	Acute Inflammation	0	1
C	(Unremarkable for above)	0/59	0/59
D	(Unremarkable for above)	0/50	0/48
E	(Unremarkable for above)	0/127	0/126
	88-Week Dietary Studies		
F	Mononuclear infiltration	5/34	6/26
	104-Week Dietary Studies		
G	Lymphocytic thyroiditis	0/100	2/100
H	Thyroiditis	0/70	1/69
I	Mononuclear infiltration	3/95	5/98
J	Mononuclear infiltration	3/48	4/45
K	(Unremarkable for above)	0/48	0/50

Additionally, the registrant has provided comments on the incidence of thyroiditis in the propanil chronic mouse study as presented below:

"An increased incidence of thyroiditis was observed in the Group 6 (180 ppm, 85.4% a.i. Lot 9287) female mice in the 24-month dietary oncogenicity study of Stam(R) technical herbicide, (Hazleton Laboratories America, Project No. 417-400, Rohm and Haas Report No. 82RC-68). Incidences in all other dose groups (5, 30, and 180 ppm, 98% Lot LSPP3-0031R) were comparable to controls, as is seen in Table 1 attached.

"The report concluded that the possibility of a 'relationship between Stam technical (85.4% purity) and thyroiditis could not be excluded.'

"As can be seen in Table 1, the incidence of thyroiditis in all the groups is low, and the apparent difference in incidence in Group 6 is attribut[e]able to 8/67 females with a severity grade of minimal, a very small degree of change. All of these were among the survivors killed at term (105 weeks, report p. 495). Inflammatory lesions of various types are normally seen in aged mice, as is illustrated by the thyroid historical control information included as item #2 of this Supplement. No other indications of toxicity to the thyroid were noted in this study or at these and higher dose levels in other studies of Stam technical.

"For these reasons, and noting that the lesion could not be unequivocally related to treatment, this finding is considered not to be due to the administration of Stam Technical.

"Since this was the only parameter examined in this study in which a change was noted with the 85.4% technical which was not observed with the 98% technical, it was concluded that no biologically significant differences were demonstrated between the 98% and 85.4% purity technical lots within the context of this two year study." [End of quotation.]

TB concurs with the registrant's position that the occurrence of thyroiditis in group 6 females is not compound-related.

The grades of the lesions of thyroiditis in group 6 females included <u>one</u> grade of moderate (out of 9 incidences), in addition to eight grades of minimal. The percentage incidence of thyroiditis in group 6 females is 13.4% (9/67) and is within the range of 0 to 23% for thyroiditis in the historical controls at 88 weeks.

The incidence of thyroditis in group 6 females exceeds the range of historical controls of the 104-week studies. However, since the 88-week time period occurs <u>before</u> the 104th week in a study, the occurrence of 23% thyroiditis at 88 weeks can be applied to the propanil mouse study.

<u>Issue #3</u>

The registrant has submitted the CSF for the technicals used in the major toxicology studies.

The CSF of the 85.4% and 98% technicals used in the 2-year mouse study were also submitted. The following additional studies used the 85.4% technical: rat and rabbit teratology studies. The 97% technical was used in the 2-year dog, 2-year rat, and 3-generation rat reproduction studies. As published in TAP 23, 650-659 (1972), the 97% technical contained 97% propanil, 0.9% dichloroaniline, and 2% unknowns.

The fact that the 85.4% technical contained [redacted] "total other impurities"* may be significant in light of the bilateral retinal degeneration in males in group 6 of the propanil mouse study.

There are two technicals presently registered with the Agency. A 98% technical product registered in April 1985 and an 85-88% technical product registered since 1972. Additionally, a 97% purity technical was used in the 1960s.

The 2-year rat feeding study, the 2-year dog feeding study, the 3-generation rat reproduction study, the rat metabolism study, and the 90-day rat feeding study were all done in the 1960s by the Medical College of Virginia using a 97% purity technical, which may be chemically different from both the 98% and 85-88% technicals used since that time.

The rat and rabbit teratology studies were performed with the 85.4% purity technical and were performed in 1980. There are no teratology studies with the 98% technical.

The mutagenicity studies were performed with either the 85-88% technical or the 98% technical.

The 2-year mouse oncogenicity study was performed with both the 98% technical (doses of 5, 30, and 180 ppm) and the 85.4% technical (dose of 180 ppm).

It is uncertain whether two complete toxicology data bases are required for the two different presently registered technicals or whether the toxicology data base can be comprised of studies using either of the two technicals (which may contain significant differences in impurities). Depending upon the technical registrations that the registrant wishes to maintain and the type and quantity of impurities therein, it may be necessary to repeat certain studies, for example, the reproduction study (using 85-88% and/or 98% technicals), the teratology studies (using 98% technical), the mouse oncogenicity

*These impurities are [redacted]

study (with 85-88% technicals), and possibly other studies. The registrant is required to address this issue in satisfactory detail.

Additional Issue

The HDT in the 2-year mouse oncogenicity study may not have been the MTD. The HDT was 180 ppm and was based on the results of a 90-day range-finding study in mice. In the range-finding study, severe toxic effects were produced at 1600 and 12,800 ppm. The effects observed at 200 ppm were hepatocytic pleomorphism in 3/10 females and multifocal hepatocellular necrosis in 1/10 males. The HDT for the 2-year mouse study was chosen to be 180 ppm, which is approximately eight times lower than the dose level of 1600 ppm, where "life-threatening" toxic effects were observed. TB considers the 180 ppm level for the 2-year mouse study to be probably too low and to be based only on hepatocytic pleomorphism and multifocal hepatocellular necrosis, which may be insufficient toxic endpoints. The registrant is requested to provide an explanation and/or rationale as to why the HDT in this study (180 ppm) might be considered the MTD.

~~Attachments~~