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

005507

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

Subject: Terbutryn Registration Standard

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A registration standard on terbutryn was written by Alex Arce. Toxicology Branch has found it necessary to modify that standard. The following registration standard on terbutryn should be used in place of the earlier one.

080813

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Registration Standard for Terbutryn

Toxicology Chapter

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A. Toxicology Profile

81 Series Acute Toxicity and Irritation Studies

81-1 Acute Oral

An acute oral LD_{50} of 1.9 g/kg in males and 2.1 g/kg in females has been reported for technical terbutryn in Charles River rats (MRID 00045432).

Toxicity Category: ~~II~~ III gwh
Core Classification: Minimum

This study satisfies the requirement for acute oral toxicity.

81-2 Acute Dermal

An acute dermal LD_{50} of >20.0 gm/kg for technical terbutryn has been reported for albino rabbits (MRID 00048742).

Toxicity Category: ~~III~~ IV gwh
Core Classification: Minimum

This study satisfies the requirement for acute dermal toxicity.

81-3 Acute Inhalation

An acute inhalation study was not found on technical terbutryn. An acute inhalation toxicity study on the technical product is required for registration.

81-4 Primary Eye Irritation

An adequate primary irritation test on technical terbutryn was not available. However, a test was available on a product containing 76% terbutryn (MRID 00146728).

Toxicity Category: III
Core Classification: Guideline

This study satisfies the requirement for a primary eye irritation study, since this test is not required on the technical product.

81-5 Primary Dermal Irritation

A primary skin irritation test (MRID 00048741) was reported for technical terbutryn in albino rabbits. The P.I. = 0. No irritation occured at 72 hours.

Toxicity Category: IV
Core Classification: Minimum

This study satisfies the requirement for primary skin irritation.

81-6 Dermal Sensitization

A study for technical terbutryn on dermal sensitization was not found. A dermal sensitization study is available on a product which contains 76% terbutryn (MRID 00146730). The results indicate that terbutryn should not be classified as a skin sensitizer in guinea pigs.

Core Classification : Minimum

A dermal sentization study on technical terbutryn is not required.

81-7 Acute Delayed Neurotoxicity

Terbutryn is not structurally related to a known group of cholinesterase inhibitors, thus acute delayed neurotoxicity testing is not required.

82 Series Subchronic Testing

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82-1 Subchronic Oral

Valid subchronic oral toxicity studies are not available on technical terbutryn; however, technical terbutryn has been tested for chronic toxicity in the rat and the dog. The six month dog study satisfies the requirement for subchronic toxicity testing. See section 83-1 for further requirements for toxicity testing in the rat.

82-2 Subchronic Dermal (21-day)

Technical terbutryn has not been evaluated in an 21-day subchronic dermal study and, thus the subchronic dermal hazard of the product cannot be evaluated. A 21-day dermal study has been submitted but has been invalidated (MRID 00085245).

A 21-day dermal study in the rabbit is required.

82-3 Subchronic Dermal (90-day)

Extended dermal exposure to technical terbutryn is not expected to occur and, thus testing in a 90-day dermal study is not required.

82-4 Subchronic Inhalation

An LD_{50} for inhalation toxicity has not been determined for technical terbutryn. The necessity for a subchronic inhalation study must be reserved until a LD_{50} is determined.

82-5 Subchronic Neurotoxicity

Terbutryn is not structurally related to a known group of cholinesterase inhibitors, thus acute delayed neurotoxicity testing is not required.

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83 Series Chronic and Long Term Studies

83-1 Chronic Toxicity

a. Rodent

A two year rat feeding study (MRID 00035923) is available, in which rats received technical terbutryn in their diet at levels of 0, 2, 300 and 3000 ppm. Decreased body weight gain for both males and females and an increased incidence of focal cytomegaly in livers of female rats was seen at 3000 ppm. Complete hematology and clinical chemistry according to guideline standards was not done in this study and thus a NOEL for these parameters could not be determined.

Although a complete two year toxicity study with histopathology will not be required, a well-designed 24-month feeding study will be required for registration with emphasis on hematology and clinical chemistry parameters in order to determine a NOEL for these effects.

Core Classification: Supplementary

b. Non-rodent

A six-month beagle dog study (MRID 00029152) is available, in which dogs received technical terbutryn at levels of 0, 10, 25 and 50 mg/kg/day in the diet. At 25 and 50 mg/kg/day mucosal thickening of various segments of the small intestine and submucosal lymphoid hyperplasia in the pyloric region of the stomach was observed. The NOEL was 10 mg/kg/day.

Core Classification: Minimum

This study was submitted during the period of time that six-month dog toxicity studies were considered and accepted as chronic studies, thus the requirement for chronic toxicity testng in a non-rodent has been satisfied.

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83-2 Oncogenicity

a. Mouse

A two year carcinogenicity study in Charles River CD-1 mice (MRID 00029153) is available, in which technical terbutryn was administered in the diet at levels of 0, 3, 1000, and 3000 ppm. No evidence of oncogenicity was observed for terbutryn in this study.

Core Classification: Minimum

The requirement for oncogenicity testing in the mouse has been satisfied.

b. Rat

A two year chronic toxicity study in CD rats (MRID 00035923) is available in which the oncogenic potential of technical terbutryn was studied. At 3000 ppm in the diet terbutryn induced a statistically significant increase in the number of mammary tumor bearing female rats, in combined hepatocellular adenomas and carcinomas in female rats, in combined thyroid follicular cell adenomas and carcinomas in male rats and in testicular interstitial cell adenomas

in male rats.

Core Classification: Minimum

The requirement for oncogenicity testing in the rat has been satisfied.

83-3 Teratogencity

Teratology studies have been conducted on terbutryn in rats (MRID 00152764) and rabbits (MRID 00152763). Terbutryn was not teratogenic to either species. The NOEL for maternal toxicity in the rabbit was 10 mg/kg and 50 mg/kg for the rat, based upon decreased food consumption, increased food [illegible] decreased body weight gain and stool changes in rabbits at 50 mg/kg and incr[illegible] mortality, salivation, urine staining, blood discharge and weight loss at 500 mg/kg in the rat. The NOEL for fetotoxicity in the rat and rabbit was 50 mg/kg based upon reduced ossification and misalignment of the sternebrae and centrum vertebrae, reduced ossification of the metatarpals, proximal phalanges and distal phalanges of the forepaw and reduced ossification of the metacarpals and distal phalanges of the hindpaw in rats at 500 mg/kg and reduced ossification of sternebrae in rabbits at 75 mg/kg.

Core Classification (both studies). Minimum

The teratogencity testing requirements for terbutryn have been satisfied.

83-4 Reproduction

A three generation reproduction study in Charles River CD rats (MRID 00035659) is available. Terbutryn was administered in the diet at levels of 2, 300 and 3000 ppm. At 3000 ppm, decreased mean body weights and food consumption values were found for the F_0, F_1 and F_2 parents as well as decreased pup weights in all generations at lactation day 21.

Core Classification: Minimum

This study satisfies the requirement for a reproduction study i rodents.

84 Series Mutagenicity

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84-2 Mutagenicity Tests

The requirements for mutagenicity testing have not been satisfied. Two submitted studies were classified Supplementary data (host mediated rat: Ciba-Geigy No. 810171 of November 20, 1981; MRID 00100653 and Mutagenicity chromosome studies in germinal epithelium of male mice, No. 810169 of November 23, 1981; MRID 00100655). One chromosomal study (in-vivo cytogenetics in hamster; MRID 00100654) was acceptable and indicated no mutagenic activity. Terbutryn has also been tested in the nucleus anomaly test (Chinese hamster; MRID 00157846). The nucleus anomaly test was negative but was found to be an unacceptable assay. The following mutagenicity studies will be required for the registration of terbutryn in order to better define the mechanism of tumor induction of terbutryn in rats:

1. in-vitro mammalian gene mutation assay;

2. a chromosome aberration study in rats;
3. unscheduled DNA synthesis assay in rat hepatocytes; and
4. a repeat of the sister chromatid assay in rats.

These assays are requested to satisfy guideline requirements and also to provide more information on the mechanism of tumor production in the rat. The need for additional mutagenicity data was also suggested by the Peer Review Committee of Toxicology Branch to aid in determining the classification of terbutryn as an oncogen.

85 Series Special Studies

85-1 Metabolism

Metabolism studies (MRID 00100640) have been conducted in the rat using both ring and methylthio-^{14}C-labelled terbutryn. Eighty-five percent of the ring-labelled dose was excreted within 72 hours in urine and feces. Sixty-two percent of ^{14}C-methylthio-labelled terburtryn was recovered with 72 hours in expired CO_2. The major pathways for metabolism are desulfuration, N-deethylation and S-demethylation.

These studies only partially meet requirements for a metabolism study on terbutryn. A metabolism study will be required for the registration of terbutryn that meets present guideline standards (i.e. multiple dosing).

85-2 Domestic Animal Safety

There are no data available to assess the animal safety of the product. However, terbutryn is not intended for animal uses and the chances of contamination are negligible. No studies are required on domestic animal safety.

85-3 Dermal Absorption

Two studies on dermal absorption of terbutryn in rats have been completed. The first study (MRID 00100656) was found to be unacceptable. In the second study (MRID not assigned), significant quantities of terbutryn were absorbed dermally at all doses and time intervals. Representative percent absorptions for 10 hours exposure were 22.02, 4.44 and 2.29% for doses of 0.05, 0.5 and 5.0 mg/10cm^2. Animals washed at ten hours and maintained for an additional 48 hours showed total absorptions of 34.48, 12.26 and 8.25 percent of the respective doses.

Core Classification: Acceptable

Further studies on dermal absorption are not required.

Information on Human Effects

No data related to human effects are available. The product is not expected to induce sensitization, allergic skin reactions or photosensitization. Data is not required.

Data Gaps

Terbutryn is registered for postemergence use on winter wheat and barley; preemergence and preplant incorporated on grain sorghum; and preemergence and postemergeence on fallow and has food tolerances. The following Guideline toxicology studies can be required for registration.

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81-1 Acute Oral Toxicity
81-2 Acute Dermal Toxicity
81-3 Acute Inhalation Toxicity
81-4 Primary Eye Irritation
81-5 Primary Dermal Irritation
81-6 Dermal Sensitization
81-7 Acute Delayed Neurotoxicity

82-1 Subchronic Oral Dosing
in two species
82-2 Subchronic Dermal (21-day)
82-3 Subchronic Dermal (90-day)
82-4 Subchronic Inhalation

83-1 Chronic Toxicity
83-2 Oncogenicity
in two species
83-3 Teratogenicity
in two species
83-4 Reproduction

84-2 Mutagenicity

85-1 Metabolism
85-3 Dermal Absorption

Based on this assessment of the toxicology data base for Terbutryn the following Guideline Toxicology studies have been identified as data gaps and are required.

81-3 Acute Inhalation Toxicity on Technical Terbutryn
82-2 Subchronic Dermal (21-day)
84-1 Mutagenicity

This data requirement is only partially satisfied and additional testing is required for:

In-vitro mammalian gene mutation assay;
A chromosome aberration study in rats;
Unscheduled DNA synthesis assay in rat hepatocytes; and
A repeat of the sister chromatid exchange assay in rats.

85-1 Metabolism

This data requirement is only partially satisfied and additional testing is required to meet 1982 EPA Guidelines for a metabolism study (i.e. multiple dosing).

Based on this assessment of the toxicology data base for Terbutryn the following additional non-guideline study is required.

Special Studies

The registrant is required to design and perform a 24 month study in rats to determine the no-observed-effect level (NOEL) for the effects of terbutryn on

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matology and clinical chemistry parameters. A NOEL was not determined for these fects in a two year chronic/oncogenicity study due to deficiencies in study design.

Tolerances and Tolerance Reassessment

Tolerances for terbutryn have been approved for the RACs listed below.

blished Tolerances

op	Tolerance	Food Factor	mg/day(1.5kg)
rley	0.100 ppm	0.03	0.000045
rghum	0.100 ppm	0.03	0.000045
eat	0.100 ppm	10.36	0.015540

The ADI for terbutryn was based on a six-month dog feeding study. The no-served-effect level (NOEL) for this study was 10 mg/kg/day based upon mucosal ickening of various segments of the small intestine and submucosal lymphoid perplasia in the pyloric region of the stomach seen at 25 and 50 mg/kg/day. ing a safety factor of 1000, since a NOEL was not determined in the chronic study, a RADI of 0.0100 mg/kg/day can be calculated. This is ivalent to a I of 0.6 mg/day for a 60 kg individual. The TMRC of terbutryn in the da ly et based on the total tolerances above and a daily food intake of 1.5 kg is 000260 mg/kg/day. Under these conditions 2.6050% of the ADI has been utilized.

Toxicological Issues

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Oncogenicity in the Rat

When administered in the diet to Charles River CD rats terbutryn induced a tistically significant increase in combined mammary gland tumors (adenomas, roadenomas and adenocarcinomas) and combined hepatocellular adenomas and cinomas in females and in combined thyroid follicular adenomas and carcinomas testicular interstitial cell alenomas in males at the highest dose tested 00 ppm). These data were presented to the Toxicology Branch Peer Review mittee. Their conclusions, as drawn from their report, were as follows:

e Committee concurred that the classification of terbutryn, considering all of available information, should be category C since the chemical produced 1) a ginal response in a tissue (mammary gland) known to have a high and variable kground rate, 2) an increase in combined benign and malignant tumors (testicular, roid and liver) with an agent showing no response in a variety of short-term ts for mutagenicity (limited, but negative mutagenicity data were available). Committee also considered a category B-2 classification for terbutryn since ors were produced at multiple sites and since positive, but not conclusive, ucture activity relationship (SAR) data were available. The SAR data was not sidered conclusive since, for propazine, historical control data on the mammary ors seen in the study is still outstanding and, for atrazine, only a preliminary ort on the incidence of mammary tumors was available and the final report has been evaluated. In addition four thyroid inhibitors which are structurally ilar to terbutryn, are known to induce thyroid neoplasia. The Committee felt t a category C classification was most appropriate but that positive information the area of mutagenicity for terbutryn and/or mutagenicity and oncogenicity other structurally related triazines could raise terbutryn to category B-2 ssification. In light of this possibility, the Committe decided that a titative estimation of the oncogenic potential for humans should be developed."

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Additional mutagnicity data has been requested of the Registrant in this egistration Standard. A quantitative estimation of the oncogenic potential has een completed by Toxicology Branch.

		[Q^* $(ppm)^{-1}$)]
Females		
Mammary:	adenomas + carcinomas	.89 x 10^{-4}
	adenomas + carcinomas + fibroadenomas	2.27 x 10^{-4}
Liver:	adenomas	1.09 x 10^{-4}
	adenomas + carcinomas	1.33 x 10^{-4}
Males		
Thyroid:	adenomas	5.12 x 10^{-4}
	adenomas + carcinomas	8.70 x 10^{-4}
Testes:	adenomas	1.68 x 10^{-11}

The appropriate Q^* to use for risk assessment is that which gives the most nservative estimate of risk. In this case it would be that derived from thyroid mors seen in male rats: 8.70 x 10^{-4} (ppm^{-1}). This would correspond to x 10^{-2} $(mg/kg/day)^{-1}$ for human assuming 60 kg average body weight for humans i doing a body surface correction when converting from rat to human.

The upper 95% confidence limits on the dietary oncogenic risks for terbutryn ossible human oncogen) is 2.6 x 10^{-5}. This is based on barley, sorhhum and eat at tolerance levels of 0.1 ppm.

TABLE A
GENERIC DATA REQUIREMENTS FOR TERBUTRYN

Data Requirement	Composition 1/	Use Patterns 2/	Does EPA Have Data To Satisfy This Requirement? (Yes, No or Partially)	Bibliographic Citation	Must Additional Data Be Submitted Under FIFRA Section 3(c)(2)(B)?3/
§158.135 Toxicology					
ACUTE TESTING:					
81-1 - Acute Oral - Rat	TGAI	A, B	Yes	00045432	No
81-2 - Acute Dermal - Rabbit	TGAI	A, B	Yes	00048742	No
81-3 - Acute Inhalation - Rat	TGAI	A, B	No		Yes
81-4 - Eye Irritation - Rabbit	TGAI	A, B	Yes	00146728	No
81-5 - Dermal Irritation - Rabbit	TGAI	A, B	Yes	00048741	No
81-6 - Dermal Sensitization - Guinea Pig	TGAI	A, B	Yes	00146730	No
81-7 - Acute Delayed Neurotoxicity - Hen	TGAI	A, B	No4/		No
SUBCHRONIC TESTING:					
82-1 - 90-Day Feeding -					
Rodent	TGAI	A, B	Yes	00035923	No
Non-rodent	TGAI	A, B	Yes	00029152	No

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Data Requirement	Composition 1/	Use Pattern 2/	Does EPA Have Data To Satisfy This Requirement? (Yes, No or Partially)?	Bibliographic Citation	Must Additional Data Be Submitted Under FIFRA Section 3(c)(2)(B)? 3/
§158.135 Toxicology (Cont.)					
82-2 - 21-Day Dermal-	TGAI	A, B	No		Yes
82-3 - 90-Day Dermal-	TGAI	A, B	No 5/		No
82-4 - 90-Day Inhalation -	TGAI	A, B	No		No
82-5 - 90-Day Neurotoxicity-	TGAI	A, B	No 4/		No
CHRONIC TESTING:					
83-1 - Chronic Toxicity					
Rodent	TGAI	A, B	Partially 6/	000 5923	Yes
Non-rodent	TGAI	A, B	Yes	00029152	No
83-2 - Oncogenicity Study -					
Rat	TGAI	A, B	Yes	00035923	No
Mouse	TGAI	A, B	Yes	00029153	No
83-3 - Teratogenicity -					
Rat	TGAI	A, B	Yes	00152764	No
Rabbit	TGAI	A, B	Yes	00152763	No
83-4 - Reproduction - Rat	TGAI	A, B	Yes	00035659	No

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TABLE A
GENERIC DATA REQUIREMENTS FOR TERBUTRYN

Data Requirement	Composition 1/	Use Pattern 2/	Does EPA Have Data To Satisfy This Requirement? (Yes, No or Partially)	Bibliographic Citation	Must Additional Data Be Submitted Under FIFRA Section 3(c)(2)(B)? 3/
§158.135 Toxicology (continued)					
MUTAGENICITY TESTING					
84-2 - Gene Mutation	TGAI	A, B	Partially 7/	00157845	Yes
84-2 - Chromosomal Aberration	TGAI	A, B	Partially 7/	00100654	Yes
84-2 - Other Mechanisms of Mutagenicity	TGAI	A, B	No 7/		Yes
SPECIAL TESTING					
85-1 - General Metabolism	PAI or PAIRA	A, B	Partially 8/	00100640	Yes
85-2 - Domestic Animal Safety	Choice	A, B	No		No
85-3 - Dermal Absorption	TGAI	A, B	Yes	not assigned	No
85-4 - Special Study	TGAI	A, B	No 6/		Yes

1/ Composition: TGAI Technical Grade Active Ingredient; PAI = Pure Active Ingredient; PAIRA = Pure Active Ingredient, Radiolabelled; Choice = Choice of several test substances determined on a case-by-case basis.
2/ The use patterns are coded as follows: A = Terrestrial, Food Crop; B = Terrestrial, Non-Food; C = Aquatic, Food Crop; D = Aquatic, Non-Food; E = Greenhouse, Food Crop; F = Greenhouse, Non-Food; G = Forestry; H = Domestic Outdoor; I = Indoor; IP = Industrial Preservative.
3/ Unless otherwise specified data must be submitted no later than six months after publication of this Standard.
4/ Terbutryn is not structurally related to a known group of cholinesterase inhibitors, thus neurotoxicity testing is not required.
5/ Extended dermal exposure is not expected to occur, thus subchronic dermal testing is not required.
6/ A special 24 month rat study is required to determine a no-observed-effect-level for hematology and chlinical chemisty parameters. This study must be submitted three years after publication of this Standard.

7/ The following mutagenicity assays are required on terbutryn: in-vitro mammalian gene mutation assay; a chromosome aberration assay in rats; unscheduled DNA synthesis assay in rat hepatocytes; and a repeat of a sister chromatid exchange assay in rats. These studies must be submitted one year after publication of this Standard.

8/ Additional metabolism information is necessary to meet 1982 EPA guidelines, i.e. multiple dosing phase. This study must be submitted one year after publication of this Standard.

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TABLE B

PRODUCT SPECIFIC DATA REQUIREMENTS FOR MANUFACTURING-USE PRODUCTS CONTAINING TERBUTRYN

Data Requirement	Composition 1/	Does EPA Have Data To Satisfy This Requirement? (Yes, No or Partially)	Bibliographic Citation	Must Additional Data Be Submitted Under FIFRA Section 3(c)(2)(B)? 2/
§158.135 Toxicology				
ACUTE TESTING				
81-1 - Acute Oral - Rat	MP	Yes	00146725	No
81-2 - Acute Dermal	MP	Yes	00146726	No
81-3 - Acute Inhalation - Rat	MP	Yes	00146727	No
81-4 - Primary Eye Irritation - Rabbit	MP	Yes	00146729	No
81-5 - Primary Dermal Irritation - Rabbit	MP	Yes	00146728	No
81-6 - Dermal Sensitization Guinea pig	MP	Yes	00146730	No

1/ Composition: MP = Manufacturing-use product.
2/ Unless otherwise specified data must be submitted no later than six months after publication of this Standard

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00029152	Liddell, J.A.; Hepler, D.L.; Beck, L.S.; et al. (1980) Six Month Oral Toxicity Study of Terbutryn Technical in Beagle Dogs: Project No. 1421. (Unpublished study received June 5, 1980 under 100-540; prepared by Elars Bioresearch Laboratories, Inc. submitted by Ciba-Geigy Corp., Greensboro, N.C.; CDL:242568-B)
00029153	Jessup, D.C.; Marshall, P. (1980) Two-Year Carcinogenicity Stu in Mice: IRDC No. 382-005. (Unpublished study received June 5, 1980 under 100-540; prepared by International Research and Development Corp., submitted by Ciba-Geigy Corp., Greensboro, N.C.; CDL:242569-A)
00035659	Jessup, D.C., Wrenn, J.M.; Rajasekaran, D.; et al. (1980) Three Generation Reproduction Study in Rats: IRDC No. 382-011. (Unpublished study received June 5, 1980 under 100-540; prepared by International Research and Development Corp., submitted by Ciba-Geigy Corp., Greensboro, N.C.; CDL:242572-A)
00035923	Jessup, D.C.; Gunderson, G.; Ferrell, J.F.; et al. (1980) 2-Year Chronic Oral Toxicity Study in Rats: IRDC No. 382-008. Rev. (Unpublished study received June 5, 1980 under 100-540; prepared by International Research and Development Corp. in cooperation with Experimental Pathology Laboratories, Inc. submitted by Ciba-Geigy Corp., Greensboro, N.C.; CDL:242570-A)
00045432	Mastri, C. (1970) Report to Geigy Agricultural Chemicals: Acute Oral Toxicity Study on GS-13529 Technical and GS-14260 Technical in Male and Female Albino Rats: IBT No. A8087. (Unpublished study including submitter summary, received April 21, 1971 under 1F1154; prepared by Industrial Bio-Test Laboratories, Inc. submitted by Ciba-Geigy Corp., Ardsley N.Y.; CDL:093472-F)
00048741	Dean, W.P. (1977) Primary Skin Irritation Study in Rabbits: IRDC No. 382-021. (Unpublished study received August 16, 1977 under 100-496; prepared by International Research and Development Corp., submitted by Ciba-Geigy Corp., Greensboro, N.C.; CDL: 231313-M)
00048742	Dean, W.P. (1977) Acute Dermal Toxicity Study in Rabbits: IRDC No. 382-021. (Unpublished study received August 16, 1977 under 100-496; prepared by International Research and Development Corp., submitted by Ciba-Geigy Corp., Greensboro, N.C.; CDL: 231313-N)
00085425	Mastri, C. (1967) Report to Geigy Agricultrual Chemicals, Division of Geigy Chemical Corporation: 21-day Subacute Dermal Toxicity of GS-14260 80W: IBT No. A5456. (Unpublished study received January 20, 1968 under 8F0714; prepared by Industrial Bio-Test Laboratories, Inc., submitted by Geigy Chemical Co., Ardsley. N.Y.; CDL:091235-G)

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00100640 Larsen, G.: Bakek, J.; Feil, V. 1978. Metabolism of ^{14}C-Terbutryn by Rats and Goat. Source unknown ?:? (Also in unpublished submission received May 4, 1982 under 100-496; submitted by Ciba-Geigy Corp.; CDL:247365-C)

00100653 Arni, P. (1981) Intrasanguine Host-Mediated Assay with S. Typhimurium: No. of experiment: 810171. (Unpublished study received May 4, 1982 under 100-540; prepared by Ciba-Geigy Ltd., Switzerland, submitted by Ciba-Geigy Corp., Greensboro, N.C.; CDL:247368-B)

00100654 Hool, G.; Puri, E. (1981) Chromosome Studies in Somatic Cells, Bone Marrow, GS 14260, Chinese Hamster--Dosage: 750, 1500, 3000 mg/kg: No.of experiment: 810173. (Unpublished study received May 4, 1982 under 100-540; prepared by Ciba-Geigy Ltd., Switzerland, submitted by Ciba-Geigy Corp., Greensboro, N.C. DCL:247368-C)

00100655 Hool, G. (1981) Chromosome studies in Male Germinal Epithelium, Spermatogonia GS 14260, Mouse--Dosage: 486, 1458 mg/kg; No. of experiment: 810169. (Unpublished study received May 4, 1982 under 100-540; prepared by Ciba-Geigy Ltd., Switzerland, submitted by Ciba-Geigy corp., Greensboro, N.C.; CDL:247368-D)

00100656 Simoneaux, B. (1982) Dermal Absorption of S-Triazine-^{14}C-Terbutryn by Rats: M5-10-9A: Report No. ABR-82016. (Unpublished study received May 4, 1982 under 100-540, submitted by Ciba-Geigy Corp., Greensboro, N.C.; CDL:070914-N)

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00152763 Infurna, R.; Arthur, A. (1985) Terbutryn Technical; A Teratology Study in New Zealand White Rabbits: (MIN 842105): Report 85010. Unpublished study prepard by Ciba-Geigy Corp.

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00157346 Ciba-Geigy Ltd. (1985) Nucleus Anomaly Test in Somatic Interphase Nuclei: Gs 14260: Chinese Hamster: (Test for Mutagenic Effects on Bone Marrow Cells): Report No. 810174. Unpublished study.

Murphy, T.; and Simoneaux, B.; Dermal Absorption of Terbutryn in Rats. Biochemistry Dept., Agricultural Division, Ciba-Geigy, Report No. ABR-86017.

File Last Updated _____ Current Date

Study/Lab/Study #/Date	Material	EPA Accession No.	Results: LD_{50}, LC_{50}, PIS, NOEL, LEL	Guideline No.
Registration standard Teratology - rat; Industrial Bio-Test; #B-904; May 11, 1972	Terbutryn Technical		Teratogenic NOEL > 50 mg/kg/day Fetotoxic NOEL > 50 mg/kg/day (highest level tested). Levels tested = 0, 25, and 50 mg/kg.	00550L 005507 Minimum 001207 001208
Teratology - rat; Ciba-Geigy; #2271-1100 January 26, 1972	Terbutryn Technical		Teratogenic NOEL > 250 mg/kg/day (HDT). Fetotoxic NOEL = 25 mg/kg/day. Fetotoxic LEL = 100 mg/kg/day (increased number of resorptions). Levels tested = 0, 25, 100, and 250 mg/kg.	Minimum
3-Generation reproduction - rat; IRDC; # #382-011; October 8, 1979	Terbutryn Technical	242568 242569 242570 242571 242572 242573	NOEL = 300 ppm. LEL = 3000 ppm (HDT) (decreased body weight gain and fertility indices in both males and females). Levels tested = 0, 2, 300, and 3000 ppm in Charles River strain.	Minimum 001204
6-Month feeding - dog; Elars Bioresearch Labs; #1421; January 22, 1980	Terbutryn Technical Batch # FL-761552	242568 242569 242570 242571 242572 242573	NOEL = 10 mg/kg/day (lowest level tested). LEL = 25 mg/kg/day (increased salivation, agitation). Temporary posterior ataxia in highest level tested (50 mg/kg/day). Levels tested = 0, 10, 25, and 50 mg/kg in Beagles.	Minimum 001204
90-Day feeding - dog; Industrial Bio-Test; #T-5286; November 13, 1967	Terbutryn Technical		Invalid by Canadian Review.	001205 001206

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Study/Lab/Study #/Date	Material	Accession No.	Results: LD_{50}, LC_{50}, PIS, NOEL, LEL	TOX Category	CORE Grade/ Doc. No.
4-Week feeding - mice; IRDC; #382-002; September 17, 1976 Range-finding Study	Terbutryn Technical		NOEL = 3000 ppm. LEL = 10,000 ppm (decreased body weight gain, corneal opacity, eccentric pupils). Levels tested = 0, 30, 100, 300, 600, 1000, 3000, 10,000, and 30,000 ppm.		Minimum 001208
2-Year feeding/oncogenic - rat; IRDC;382-008; March 27, 1980	Terbutryn Technical Batch # FL-761552	242568 242569 242570 242571 242572 242573	Oncogenic NOEL = 300 ppm. Oncogenic LEL = 3000 ppm (significant increase in thyroid follicular adenomas, mammary gland adenoma, and liver adenoma). Borderline statistical significance in testicular interstitial cell adenoma in males. Systemic NOEL = 300 ppm. Systemic LEL = 3000 ppm (body weight decrease, blood biochemical changes). Dose levels tested in Charles River strain - 0, 2, 300, and 3000 ppm.		Minimum 001204
2-Year Oncogenic - mice; IRDC; #382-005; April 21, 1980	Terbutryn Technical Batch # FL-761552	242568 242569 242570 242571 242572 242573	Oncogenic NOEL > 3000 ppm (HDT). Levels tested = 0, 3, 1000, and 3000 ppm in Charles River strain.		Minimum 001204
21-Day dermal - rabbit; Industrial Bio-Test; #A5456; #A5456; August 17, 1967	GS-14260 80W (Terbutryn WP)		Invalid by Canadian Review.		Invalid 001210
Mutagenic - host-mediated - rat; Ciba-Geigy; November 20, 1981	Terbutryn Technical	247365 247366 247367 247368	Negative Mutagen. No mutagenic effects at 500, 1000, or 2000 mg/kg. Given by gavage.		Supplementary 002021

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Study/Lab/Study #/Date	Material	Accession No.	Results: LD_{50}, LC_{50}, PIS, NOEL, LEL	TOX Category	CORE Grade/ Doc. No.
Mutagenic - in vivo cytogenetics test - hamster; Ciba-Geigy; Basle; Zurtzyer; #810173; November 24, 1981	Terbutryn Technical	247365 247366 247367 247368	Negative Mutagen. No chromosome aberrations in bone marrow cells at 750, 1500, or 3000 mg/kg/day given orally for 2 days.		Minimum 002021
Mutagenic - chromosome studies in germinal epithelium of male mice; Ciba-Geigy; #810169; Switzerland; November 23, 1981	Terbutryn Technical	247365 247366 247367 247368	Negative Mutagen. No chromosome aberrations in spermatogonia at 486 and 1458 mg/kg/day given by gavage for 5 days.		Supplementary 002021
Dermal absorption - rat; Ciba-Geigy; Greensboro, NC	^{14}C-Terbutryn Labeled in Triazine Ring	247365 247366 247367 247368	Inadequately reported results.		Invalid 002021
Acute oral LD_{50} - rat	TECH	III	2.5 gm/kg LD_{50} = 2.5 g/kg.	III	Minimum
Acute oral LD_{50} - mice	Terbutryn 50% WP		LD_{50} = 3.36 g/kg.	III	001206
Acute oral LD_{50} - guinea pig	Terbutryn 50% WP		LD_{50} = 0.5 g/kg.	II	001206
Acute oral LD_{50} - rabbit	Terbutryn 50% WP		LD_{50} = 0.7 g/kg.	III	001206
Acute oral LD_{50} - hen	Terbutryn 50% WP		LD_{50} = 4.g/kg.	III	001206
Acute oral LD_{50} - rat; Industrial Bio-Test; #A-8087; January 23, 1970	GS-14260 (Terbutryn) Technical		Acute Oral LD_{50}: (Canadian Review) Male: 1.9 g/kg Female: 2.1 g/kg		001209

005307

Study/Lab/Study #/Date	Material	Accession No.	Results: LD_{50}, LC_{50}, PIS, NOEL, LEL	TOX Category	CORE Grade/ Doc. No.
Acute dust inhalation - rat; Industrial Bio-Test; #N-5457 August 17, 1967	GS-14260 80W (Terbutryn WP)		Invalid by Canadian Review. LC_{50} > 8 mg/L (highest level tested).	III	Minimum 001208 001211 001212
Acute dermal LD_{50} - rabbit; Industrial Bio-Test; #A-5458 June 28, 1967	GS-14260 80W (Terbutryn) WP) 80-8% ai 50% w/v aqueous dilution		LEL = 6.8 g/kg* (NOEL not established) pale red erythema, mild edema. LD_{50} > 10.2 g/kg (highest level tested). *Canadian reviewers cite unreported data that indicate mild to moderate erythema occurred at 4.5 g/kg.	III	Minimum 001208 001213
Acute oral LD_{50} - rat; Industrial Bio-Test; #A-5458; June 28, 1957	GS-14260 80W 80.8% ai		Acute oral LD_{50} males - 2.5 g/kg ± females - 2.5 g/kg ± 0.4.	III	Minimum 001206 001208
Primary eye irritation - rabbit; Industrial Bio-Test; #A-5458; June 28, 1967	GS-14260 80W Terbutryn WP		Corneal opacity eye irritation: moderate clear by 7 days.	II	Minimum 001206 001208
Primary dermal irritation - rabbit; Industrial Bio-Test; #N-5457; August 17, 1967	GS-14260 80W Terbutryn WP		PIS = 1.3/8 No irritation at 72 hours.	IV	Minimum 001208
Acute aerosol inhalation - rat; Industrial Bio-Test; #N-5457 August 17, 1967	GS-14260 80W Terbutryn WP		LC_{50} > 27.8 mg/L 4 hours.	IV	Minimum 001208
Risk Assessment					004079

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Study/Lab/Study #/Date	Material	No.	Results: LD_{50}, LC_{50}, PIS, NOEL, LEL	TOX Category	CORE Grade/ Doc. No.
Acute oral LD_{50} - rat; Ciba-Geigy; #842204; June 27, 1984	IGRAN® 80 WDG (76% ai)	255226	LD_{50} = 1542 mg/kg (both sexes). LD_{50} = 2206 mg/kg (males only). LD_{50} for females not determined because there was 100%-mortality in two highest dosed groups leaving too few points.	III	Minimum 004458
Acute dermal LD_{50} - rabbit; Ciba-Geigy; #842178; June 27, 1984	IGRAN® 80 WDG (76% ai)	255226	LD_{50} > 2000 mg/kg (both sexes) (Limit test, only dose tested).	III	Minimum 004458
Acute inhalation LC_{50} - rat; Ciba-Geigy; #842179; June 22, 1984	IGRAN® 80 WDG (76% ai)	255226	LC_{50} > 15.5 mg/L (nominal concentration) or 2.5 mg/L (actual concentration) (4-hour exposure).	III	Minimum 004458
Primary dermal irritation - rabbit; Ciba-Geigy; #842175	IGRAN® 80 WDG (76% ai)	255226	Non-irritating (PIS = 0.29).	IV	Guideline 004458
Primary eye irritation - rabbit; Ciba-Geigy; #842176;	IGRAN® 80 WDG (76% ai)	255226	Moderately irritating (corneal involvement and irritation cleared within 7 days after instillation of test substance).	III	Guideline 004458
Dermal sensitization - guinea pig; Stillmeadow, Inc.; June 28, 1984	IGRAN® 80 WDG (76% ai)	255226	Not a sensitizer.		Minimum 004458
Risk Assessment			The dose-response slope (Q_1^*) for the multi-stage model was determined to be 9.35×10^{-6} on a µg/kg/day basis.		004381

005507

DER's

The DER's used in this standard are attached in the following order:

1. Addendum to "Acute Oral Toxicity Study on GS-13529 Technical and GS-14260 Technical in Male and Female Albino Rats" IBT No. A8087. MRID 00045432.

2. Audit report on the study listed under #1 above.

3. Acute Dermal Toxicity Study in Rabbits: IRDC No. 382-021; MRID 00048742.

4. Primary Skin Irritation study in Rabbits: IRDC No. 382-021; MRID 00048741.

5. Acute Toxicity Studies conducted on Igran 80 WDG.

6. Addendum to DER of the two year chronic rat toxicity study on terbutryn MRID 00035923.

7. DER dated 7/25/80 on the the six month dog study (MRID 00029152), the mouse oncogenicity study (MRID 00029153), the two year chronic rat toxicity study (MRID 00035923) and the 3-generation reproduction study (MRID 00035659).

8. Addendum to DER's on teratology studies in rat and rabbits on terbutryn.

9. A Teratology Study in Rabbits (MRID 00152763).

10. A Teratology Study in Rats (MRID 00152764).

11. DER dated 6/23/82 on the rat intrasanguine host-mediated assay (MRID 00100653), in-vivo hamster cytogenetic study (MRID 00100654), chromosome studies in the germinal epithelim of male mice (MRID 00100655) and the invalid dermal absorption study (MRID 00100656).

12. DER (Draft) dated 6/25/86 on the micronucleus test in chinese hamsters, (MRID 00157846).

13. Dermal Absorption Study of Terbutryn in Rats. (MRID not assigned).

14. Draft report of the Peer Review Committee on Terbutryn.

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ADDENDUM TO DER

:emical: Terbutryn

:udy: Mastri, C. (1970) Report to Geigy Agricultural Chemicals: Acute Oral Toxicity Study on GS-13529 Technical and GS-14260 Technical in Male and Female Albino Rats: IBT No. A8087. (Unpublished study including submitter summary, recieved Apr 21, 1971 under 1F1154; prepared by Industrial Bio-Test Laboratories, Inc., submitted by Ciba-Geigy Corp., Ardsley, N.Y.)

:epared By: Judith W. Hanswirth, Ph.D. 6/25/86
Mission Support Staff
Toxicology Branch/HED

:proved By: Reto Engler, Ph.D., Chief
Mission Support Staff
Toxicology Branch/HED

scussion: This study was audited by Canadian scientists in 1979. The overall mment of the auditor was as follows: "The audit and validation of this study dicate that despite minor errors and omissions, this study can be considered be scientifically acceptable."

tails of the study can be found in the audit report which is attached. This udy can now be classified as Core Minimum data.

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Government of Canada / Gouvernement du Canada

MEMORANDUM NOTE DE SERVICE

125D Terbutryn

SECURITY CLASSIFICATION / DE SÉCURITÉ

YOUR FILE / VOTRE RÉFÉRENCE

OUR FILE / NOTRE RÉFÉRENCE

DATE
March 20, 1979

Mr. P.J. Clegg
Head, Pesticides Section
Toxicological Evaluation Division

Dr. R.M. Sharma

SUBJECT / OBJET Acute oral toxicity of Terbutryne Tech. (GS-14260) in rats

Overall Comment: The audit and validation of this study indicate that despite minor errors and omissions, this study can be considered to be scientifically acceptable.

Acute oral toxicity of Terbutryne Tech. (GS-11260) in rats:

A. Audit:

1. Report no.: IBT A8027 dated January 23, 1970.

2. Date: This study was a combination of 3 range finding studies, each initiated on Dec. 17, 19 and 29, 1969 respectively. The surviving rats were observed for 14 days.

3. Protocol: Not available in the raw data.

4. Test Material: There is no information in the raw data regarding either the shipment or receipt of the test material by IBT with the exception of a work order sheet indicating that the test material was on hand on Dec. 4, 1969.

5. Animal Suitability: The IBT report indicates that rats of Charles River Strain were used. But this can't be verified from the raw data.

6. Raw data: Raw data are available for initial and final body weights, dose/sex/group, reactions observed and mortality/group.

B. Validation & Evaluation:

1. Date: See audit.

2. Protocol: Raw data indicate that 5 rats/sex/group were intubated with the test material as 25% (w/v) aqueous suspension. The body weight of the rats ranges from 187-246 g. The volume of the dose administered ranged from 1.04 to 4.30 ml/rat. The surviving rats were observed for 14 days.

3. Test Material: See audit.

4. Animal Suitability: See audit.

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5. Personnel:

Report prepared by: Carmen Mastri
Section Head
Acute Toxicity Dept.

Report approved by: M.L. Keplinger
Manager, Toxicology
&
Otis E. Fancher
Director

Technician: Mabel M. Huck

6. Execution of the study:

a. Body weight: The individual body weight data for day 0 and day 14 as presented in the final IBT report are in agreement with the raw data with the exception of minor errors.

b. Reactions: The data as given in the final report are in agreement with the raw data with the exception of minor errors & omissions. The reaction recorded for each group were as follows:

1.4 g/kg: Hyperactivity and ruffed

2g, 3g, & 4.6/kg: Hyperactivity, diarrhea muscular weakness, ruffed fur and emaciation.

c. Mortality: The mortality data as given in the final IBT report are in agreement with the raw data with the exception of minor errors. The results were as follows:

Males: 1/5, 3/5, 4/5 & 5/5 at 1.4, 2.0 3.0 and 4.6 g/kg respectively.

Females: 0/5, 2/5, 5/5 & 5/5 at 1.4, 2.0 3.0 and 4.6 g/kg respectively.

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d. LD50:

Acute oral LD50 for male: 1.9 g/kg
Acute oral LD50 for female: 2.1 g/kg

Overall Comment: The audit and validation of this study indicate that despite minor errors and omissions, this study can be considered to be scientifically acceptable.

Review of validation performed by Drs. John A. Stone and James T. Stevens.

We agree with Drs. Stone and Stevens conclusion.

R.M. Sharma

D.J. Clegg

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005507

DER

Chemical: Terbutryn

Test Material: Technical terbutryn

Study Title: Dean, W.P. (1977) Acute Dermal Toxicity Study in Rabbits: IRDC No. 382-021. (Unpublished study received Aug. 15, 1977 under 100-496; prepared by International Research and Development Corp.), submitted by Ciba-Geigy Corp., Greensboro, N.C.

Reviewed By: Judith W. Hauswirth, Ph.D.
Mission Support Staff
Toxicology Branch/HED

Approved By: Reto Engler, Ph.D., Chief
Mission Support Staff
Toxicology Branch/HED

Conclusions: The acute dermal toxicity of terbutryn is >20,000 mg/kg.

Core Classification: Minimum
Toxicity Category: IV

Materials and Methods:

Test Species: New Zealnd rabbits, 3 males and 2 females. Their weight range was 2472-2851 grams.

Methods:

Only one dosage level was used, 20,000 mg/kg. Twenty-four hours prior to administration of the test compound, the backs of each rabbit were shaved. Animals were conditioned 7 days prior to this time. The wetting agent used was 0.9% saline, the site of application was covered with gauze and occluded with Saran Wrap for 24 hours.

Rabbits were observed for signs of clinical toxicity at 24 hours and daily thereafter for a total of 14 days. Body weights of each rabbit were recorded at initiation and at 14 days.

Results:

There were no mortalities and no signs of toxicity. All of the rabbits gained weight during the study period. One rabbit showed barely perceptible erythema on days 2 and 3.

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Discussion:

The Pesticide Assessment Guidelines Subpart F call for a minimum of ten animals, five males and five females to be used for an acute dermal toxicity study. Other deficiencies in the study are as follows: 1) body weights should have been recorded at 7 days and 2) the area of skin exposed to the test material should have been reported. Although this study does not meet the guideline, the low acute dermal toxicity of terbutryn precludes the need for the study to be repeated.

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DER

Chemical: Terbutryn

Test Material: Technical terbutryn

Study Title: Dean, W.P. (1977) Primary Skin Irritation Study in Rabbits: IRDC No. 382-021. (Unpublished study received Aug. 16, 1977 under 100-496; prepared by International Research and Development Corp.), submitted by Ciba-Geigy Corp., Greensboro, N.C.

Reviewed By: Judith W. Hauswirth, Ph.D. Judith W. Hauswirth 6/2/82
Mission Support Staff
Toxicology Branch/HED

Approved By: Reto Engler, Ph.D., Chief
Mission Support Staff
Toxicology Branch/HED

Conclusions: Technical terbutryn is non-irritating to intact and abraded skin. Primary irritation score = 0.0.

Core Classification: Minimum
Toxicity Category: IV

Materials and Methods:

Test Species: New Zealand rabbits, 3 males and 3 females were used. They weighed between 2007 to 2377 grams.

Methods: Five hundred mg of the test substance was applied to two patches on each animal, one abraded, the other not. Physiological saline was used as the wetting agent and Saran Wrap as the occlusive dressing. The sites were occluded for 24 hours and scored at 24 and 72 hours.

Scoring:

Erythema and Eschar Formation:	Value (average)
No erythema	0
Very slight erythema (barely perceptible)	1
Well defined erythema	2
Moderate to severe erythema	3
Severe erythema (beet redness) to slight eschar formation (injuries in depth)	4

Edema Formation:	
No edema	0
Very slight edema (barely perceptible)	1
Slight edema (edges of area well defined by definite raising)	2
Moderate edema (raised approximately 1.0 mm)	3
Severe edema (raised more than 1.0 mm extending beyond the area of exposure)	4

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Results:

At 24 hours one animal with abraded skin had very slight erythema. This was the only animal that showed any effect.

CASWELL FILE

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004458

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

MAY 21 1985

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: New "Me too" for use of IGRAN® 80 WDG (Terbutryn) for Weed Control. EPA Reg. No. 100-ALL, Acc. No. 255226, Tox. Chem. No. 125D

TO: Robert J. Taylor
Product Manager 25
Registration Division (TS-767C)

THRU: Jane Harris, Ph. D., Section Head JCH 4/1/85
Review Section 6
Toxicology Branch
Hazard Evaluation Division (TS-769)

FROM: Roger Gardner, Toxicologist
Review Section 6 Roger Gardner 4-1-85
Toxicology Branch
Hazard Evaluation Division (TS-769) RGfull 5/21/85

Actions Requested

Review of the six studies cited in the bibliography (See Appendix I below).

Recommendations and Conclusions

1. The six studies cited below are adequate to support the registration of IGRAN® 80 WDG.

2. The acute oral, inhalation, dermal, and eye irritation studies indicate that the formulation should be classified into Toxicity Category III with respect to the four types of toxicity The skin irritation study results place the formulation into Toxicity Category IV, and the formulation was not a skin sensitizer in guinea pigs.

APPENDIX I

Data Evaluation Records

Bibliography

Choie, D., and R. Katz. June 27, 1984. Igran® 80 WDG: Acute oral toxicity in rats. Unpublished report no. 842204 prepared by Ciba-Geigy Pharmaceuticals Research, Toxicology/Pathology Division, Chemical Toxicology Subdivision, Safety Evaluation Facility, Summit, NJ. Submitted by Ciba-Geigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

Choie, D., and R. Katz. June 27, 1984. Igran® 80 WDG: Acute ermal toxicity in rabbits. Unpublished report no. 842178 prepared by Ciba-Geigy Pharmaceuticals Research, Toxicology/Pathology Division, Chemical Toxicology Subdivision, Safety Evaluation Facility, Summit, NJ. Submitted by CibaGeigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

Breckenridge, C., and R. Katz. June 22, 1984. Igran® 80 WDG: Acute inhalation toxicity study in rats. Unpublished report no. 842179 prepared by Ciba-Geigy Pharmaceuticals Research, Toxicology/Pathology Division, Chemical Toxicology Subdivision, Safety Evaluation Facility, Summit, NJ. Submitted by Ciba-Geigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

Choie, D., and R. Katz. June 27, 1984. Igran® 80 WDG: Primary dermal irritation study in rabbits. Unpublished report no. 842175 prepared by Ciba-Geigy Pharmaceuticals Research, Toxicology/Pathology Division, Chemical Toxicology Subdivision, Safety Evaluation Facility, Summit, NJ. Submitted by Ciba-Geigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

Choie, D., and R. Katz. June 27, 1984. Igran® 80 WDG: Primary eye irritation study in rabbits. Unpublished report no. 842176 prepared by Ciba-Geigy Pharmaceuticals Research, Toxicology/Pathology Division, Chemical Toxicology Subdivision, Safety Evaluation Facility, Summit, NJ. Submitted by Ciba-Geigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

Choie, D., and R. Katz. June 28, 1984. Guinea pig sensitization: Igran® 80 WDG FL 840804. Unpublished report prepared by Stillmeadow, Inc., Houston, TX. Submitted by Ciba-Geigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

005507

DATA EVALUATION RECORD

1. CHEMICAL: Terbutryn
2-tert-butylamino-4-ethylamino-6-methylthio-s-triazine

2. TEST MATERIAL: Igran® 80 WDG (76% active ingrediant, 4% related compounds)

3. STUDY/ACTION TYPE: Acute oral toxicity - rats; ("Me too" registration).

4. STUDY IDENTIFICATION: Choie, D., and R. Katz. June 27, 1984. Igran® 80 WDG: Acute oral toxicity in rats. Unpublished report no. 842204 prepared by Ciba-Geigy Pharmaceuticals Research, Toxicology/Pathology Division, Chemical Toxicology Subdivision, Safety Evaluation Facility, Summit, NJ. Submitted by Ciba-Geigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

5. REVIEWED BY:

Name: Roger Gardner
Title: Toxicologist
Organization: Review Section 6
Toxicology Branch
Signature: Roger Gardner
Date: 4-1-85

6. APPROVED BY:

Name: Jane Harris, Ph. D.
Title: Section Head
Organization: Review Section 6
Toxicology Branch
Signature: J E Harris
Date: 4/1/85

7. CONCLUSIONS: The results of the study indicate that Igran® 80 WDG should be classified into Toxicity Category III with respect to acute oral toxicity.

Core classification: Minimum

8. MATERIALS AND METHODS

Test species: Male and female Sprague-Dawley (Crl: CDBS CD (SD) BR) rats were used. Males weighed from 200 to 234 g, and females weighed from 169 to 201 g.

Experimental procedure: Groups of 5 male and 5 female rats were given single oral doses of 500, 1000, 2000, or 4000 mg

8. MATERIALS AND METHODS (continued)

test substance per kg body weight. The test substance was suspended in water and administered by gavage. The rats were fasted overnight before treatment, and they were observed for mortality and appearance of toxicological and pharmacological signs twice daily for the 14 days that followed dosing. Surviving animals were sacrificed at the end of the observation period and necropsied. Those animals that died during the study were also necropsied. Postmortem examinations were limited to gross observations.

The LD_{50} and 95% confidence limits were calculated by probit analysis.

9. REPORTED RESULTS

Signs of toxicity noted by the authors included hypoactivity, hypotonia, salivation, and ptosis which were observed one hour after dosing. Ataxia, diarrhea, and pollakiuria were also reported to occur during the first two days following dosing. Most of the surviving animals appeared normal within three days, and none of the males given the 500 mg/kg dose exhibited toxic signs. The authors stated that all of the survivors gained weight during the 14-day observation period.

No deaths occurred in rats given the 500 mg/kg dose, while all animls receiving 4000 mg/kg and females given the 2000 mg/kg dose died. Most of the deaths were reported to occur within 5 days after treatment.

The only gross observation noted at necropsy was a dark lesion in the lung of a male rat given the 4000 mg/kg dose. No other animals were found with gross lesions according to the report.

The reported LD_{50} for both sexes was 1542 mg/kg with 95% confidence limits of 1075 to 2200 mg/kg. The LD_{50} calculated for males was 2206 with 95% confidence limits of 1202 to 7049 mg/kg. The LD_{50} was not determined for females alone because the 100% mortality in the two highest dosed groups left only two points on which to base the calculations.

None of the 5 females given the 500 mg/kg dose and 2 of the 5 given 1000 mg/kg died. The LD_{50} was estimated to be 1050 mg/kg on the basis of a graph of those two points.

10. DISCUSSION

There were adequate data to support the reported LD_{50} values.

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DATA EVALUATION RECORD

1. CHEMICAL: Terbutryn
2-tert-butylamino-4-ethylamino-6-methylthio-s-triazine

2. TEST MATERIAL: Igran® 80 WDG (76% active ingrediant, 4% related compounds)

3. STUDY/ACTION TYPE: Acute dermal toxicity - rabbits; ("Me too" registration)

4. STUDY IDENTIFICATION: Choie, D., and R. Katz. June 27, 1984. Igran® 80 WDG: Acute dermal toxicity in rabbits. Unpublished report no. 842178 prepared by Ciba-Geigy Pharmaceuticals Research, Toxicology/Pathology Division, Chemical Toxicology Subdivision, Safety Evaluation Facility, Summit, NJ. Submitted by Ciba-Geigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

5. REVIEWED BY:

Name: Roger Gardner
Title: Toxicologist
Organization: Review Section 6
Toxicology Branch
Signature: Roger Gardner
Date: 4-2-85

6. APPROVED BY:

Name: Jane Harris, Ph. D.
Title: Section head
Organization: Review Section 6
Toxicology Branch
Signature: Jane Harris
Date: 4/11/85

7. CONCLUSIONS: The results of the study indicate that Igran® 80 WDG should be classified into Toxicity Category III with respect to acute dermal toxicity.

Core classification: Guideline

8. MATERIALS AND METHODS

Test species: Male and female New Zealand White strain rabbits weighing 3.1 to 3.6 kg (males) or 3.1 to 4.0 kg (females) were used

Experimental procedure: Twenty-four hours before the beginning of the study, the rabbits were prepared by clipping their backs free of hair (approximately 10% of the body surface

8. MATERIALS AND METHODS (continued)

area). The report stated that 5 animals of each sex were used in a "limit test."

The test substance was suspended in water at a concentration of 670 mg/ml, and 3 ml/kg was applied to the prepared skin site. A total of 2010 mg/kg was applied to each rabbit.

After the application of test substance, the trunks of the test animals were wrapped with gauze which was secured with adhesive tape. These dressings were then covered with orthopedic stockinets to prevent ingestion of the test substance during the 24 hour exposure. At the end of the exposure period the dressings were removed, and the test sites were gently rinsed and wiped clean.

All animals were observed twice daily for the next 14 days for the appearance of toxic signs and mortality. The rabbits were weighed on the day of dosing and on days 7 and 14 of the observation period. Surviving rabbits were sacrificed at the end of the 14-day observation period, and gross postmortem examinations were conducted.

9. REPORTED RESULTS

The authors noted no deaths, and the only sign of compound-related effects were the flaking of the skin at test sites on two rabbits. All animals gained weight during the study.

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004459

DATA EVALUATION RECORD

1. CHEMICAL: Terbutryn
2-tert-butylamino-4-ethylamino-6-methylthio-s-triazine

2. TEST MATERIAL: Igran® 80 WDG (76% active ingrediant, 4% related compounds)

3. STUDY/ACTION TYPE: Acute inhalation toxicity - rats; ("Me too" registration)

4. STUDY IDENTIFICATION: Breckenridge, C., and R. Katz. June 22, 1984. Igran® 80 WDG: Acute inhalation toxicity study in rats. Unpublished report no. 842179 prepared by Ciba-Geigy Pharmaceuticals Research, Toxicology/Pathology Division, Chemical Toxicology Subdivision, Safety Evaluation Facility, Summit, NJ. Submitted by Ciba-Geigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

5. REVIEWED BY:

Name: Roger Gardner
Title: Toxicologist
Organization: Review Section 6
Toxicology Branch
Signature: Roger Gardner
Date: 4-1-85

6. APPROVED BY:

Name: Jane Harris, Ph. D.
Title: Section Head
Organization: Review Section 6
Toxicology Branch
Signature: J.E. Harris
Date: 4/2/85

7. CONCLUSIONS: The results of the study indicate that Igran® 80 WDG should be classified into Toxicity Category III with respect to acute inhalation toxicity.

Core classification: Minimum

8. MATERIALS AND METHODS

Test species: Male and female Sprague-Dawley rats were used. Males weighed from 299 to 357 g, and females weighed from 210 to 244 g. The animals were 8 to 11 weeks of age.

MATERIALS AND METHODS (continued)

perimental procedure: The test atmosphere was generated by Trost Air Mill. The test substance was discharged into a 0 liter test chamber through which the air flowed at a rate 60 l/min. Air concentrations and particle size distributions re measured at hourly intervals, and temperature and humidity re measured at half-hour intervals during the 4-hour exposure riod. The air concentration was determined gravimetrically ing an open-faced filter. Particle size distribution was termined with a cascade impactor. Timing of the 4-hour expo- re period was not started until 15 minutes after discharging the test substance into the test chamber began.

o groups of 5 male and 5 female rats were exposed to air ntaining an expected 0 or 2 mg/l concentration of the test bstance. The rats were placed in wire mesh cages inside e test chamber, but because of the dense test atmosphere ring the 4-hour exposure period, they were not observed. ter exposure, the animals were observed twice a day for the pearance of toxic signs and mortalities. These observations re continued for 14 days after exposure.

l rats found dead during the study or sacrificed after 14 /s were necropsied, and gross observations were noted. ecial attention was given to the eyes, nasal passages, .n, bronchi, lungs, and trachea. The lungs were weighed, fused with buffered formalin, and kept along with the ver, kidneys, and abnormally appearing tissues. No stological examinations were conducted according to the port.

REPORTED RESULTS

reported temperature and humidity for the control group mals were 78° F and 57%, respectively. Those respective ues for the treated group were 74° F and 45%. The stated centrations of test substance, as determined by gravimetric hods, ranged from 2.0 to 3.5 mg/l with an average for the our exposure period of 2.5 mg/l. The nominal concentration ss of test substance used per liters flowing through the t chamber during exposure) was reported to be 15.5 mg/l. hourly mean mass diameter of particles ranged from 2.1 to um with a geometric mean standard deviation ranging from to 2.1. Particles with an equivalent aerodynamic diameter ≤ 9 um accounted for approximately 97% of the total.

re were no mortalities observed during the study.

9. REPORTED RESULTS (continued)

The authors noted that the test animals became coated with the test substance during exposure, and when removed from the chamber they exhibited chromorhiorrhea, pollakiuria, and salivation. The signs were not seen after the 7th day following exposure. Other signs reported by the investigators included alopecia, lacrimation, soft feces, hematuria, tachypnea, and unkempt appearance. All of these signs, with the exception of one case of alopecia, disappeared after the 7th day of observation. Rats which were not exposed to the test substance did not appear to be abnormal according to the report.

Group mean body weights were comparable at days 7 and 14 of the observation period, and the animals gained weight during that time. There were also no differences noted with respect to group mean lung weights.

The only necropsy findings included a reddened thymus in one control group female and a male in the treated group.

10. DISCUSSION

There were adequate data to support the conclusions of the investigators .

DATA EVALUATION RECORD

1. CHEMICAL: Terbutryn
2-tert-butylamino-4-ethylamino-6-methylthio-s-triazine

2. TEST MATERIAL: Igran® 80 WDG (76% active ingrediant, 4% related compounds)

3. STUDY/ACTION TYPE: Dermal irritation study - rabbits; ("Me too" registration)

4. STUDY IDENTIFICATION: Choie, D., and R. Katz. June 27, 1984. Igran® 80 WDG: Primary dermal irritation study in rabbits. Unpublished report no. 842175 prepared by Ciba-Geigy Pharmaceuticals Research, Toxicology/Pathology Division, Chemical Toxicology Subdivision, Safety Evaluation Facility, Summit, NJ. Submitted by Ciba-Geigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

5. REVIEWED BY:

Name: Roger Gardner
Title: Toxicologist
Organization: Review Section 6
Toxicology Branch
Signature: Roger Gardner
Date: 9-1-85

6. APPROVED BY:

Name: Jane Harris, Ph. D.
Title: Section Head
Organization: Review Section 6
Toxicology Branch
Signature: J E Harris
Date: 9/11/85

7. CONCLUSIONS: The results of the study indicate that Igran® 80 WDG should be classified into Toxicity Category IV with respect to dermal irritation (primary irritation score = 0.29).

Core classification: Guideline

8. MATERIALS AND METHODS

Test species: Male and female New Zealand White strain rabbits weighing 3.1 to 3.9 kg (males) or 3.3 to 3.8 kg (females) were used.

8. MATERIALS AND METHODS (continued)

Experimental procedure: Twenty-four hours before the beginning of the study, the rabbits were prepared by clipping their backs free of hair. Three animals of each sex were used.

The test substance was moistened with water, and 500 mg amounts were spread over a 6 cm^2 area of intact skin on each rabbit. The test sites were then covered with gauze patches which were secured with nonirritating adhesive tape. The trunk of each animal was then covered with an orthopedic stockinet.

Four hours after application of the test substance the dressings were removed, and the test sites were rinsed and gently wiped clean. One-half to one hour later the test sites were scored for edema and erythema, and they were scored again 24, 48, and 72 hours after removal of the dressings.

Erythema and eschar formation as well as edema were scored on a 5-point scale (0-4) with a maximum possible score of 8 for any site. Scoring was done according to the following classifications:

Erythema and eschar		Edema	
No erythema	0	No edema	0
Slight erythema	1	Very slight edema	1
Well-defined erythema	2	Slight edema	2
Moderate to severe erythema	3	Moderate edema	3
Severe erythema to slight eschar formation	4	Severe edema	4

Mean irritation scores were calculated as follows:

$$\text{Mean Score} = \frac{\text{Sum of Erythema and Edema Scores of All Animals}}{(\text{Total No. Animals})\ (\text{Total No. Scoring Time Points})}$$

Based on the results the test substance was classified according to the following scale:

Mean score	Irritation rating
0.0 - 0.4	Practically not an irritant
0.5 - 3.0	Slight irritant
3.1 - 5.0	Moderate irritant
5.1 - 7.0	Severe irritant
7.1 - 8.0	Corrosive

9. REPORTED RESULTS

According to the report, all the animals gained weight during the study, and no clinical signs of toxicity were observed.

Two males and two females had slight erythema at the 24-hour observation, but there was none observed at 72 hours. No other signs of irritation were observed, and the mean scores were reported to be 0, 0.67, 0.5, and 0 at 0.5 to 1, 24, 48, and 72 hours, respectively with an overall mean irritation score of 0.29.

10. DISCUSSION

The report included adequate information to support the conclusion that terbutryn is practically non-irritating.

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DATA EVALUATION RECORD

1. CHEMICAL: Terbutryn
2-tert-butylamino-4-ethylamino-6-methylthio-s-triazine

2. TEST MATERIAL: Igran® 80 WDG (76% active ingrediant, 4% related compounds)

3. STUDY/ACTION TYPE: Eye irritation study - rabbits; ("Me too" registration)

4. STUDY IDENTIFICATION: Choie, D., and R. Katz. June 27, 1984. Igran® 80 WDG: Primary eye irritation study in rabbits. Unpublished report no. 842176 prepared by Ciba-Geigy Pharmaceuticals Research, Toxicology/Pathology Division, Chemical Toxicology Subdivision, Safety Evaluation Facility, Summit, NJ. Submitted by Ciba-Geigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

5. REVIEWED BY:

Name: Roger Gardner
Title: Toxicologist
Organization: Review Section 6
Toxicology Branch
Signature: Roger Gardner
Date: 4-1-85

6. APPROVED BY:

Name: Jane Harris, Ph. D.
Title: Section Head
Organization: Review Section 6
Toxicology Branch
Signature: Jane Harris
Date: 4/11/85

7. CONCLUSIONS: The results of the study indicate that Igran® 80 WDG should be classified into Toxicity Category III with respect to eye irritation.

Core classification: Guideline

8. MATERIALS AND METHODS

Test species: Male and female New Zealand White strain rabbits weighing 3.1 to 3.8 kg (males) or 3.1 to 3.6 kg (females) were used.

8. MATERIALS AND METHODS (continued)

Experimental procedure: Nine rabbits previously examined and found without signs of eye irritation or eye defects were used in the experiment. One-tenth of a gram of the test substance was instilled into the left eye of each rabbit, and the eyelids were gently held together for one second. Thirty seconds after the instillation, the treated eyes of 3 rabbits were washed for one minute with distilled water. Washing was started 30 seconds after treatment. The eyes of the 6 remaining rabbits were washed in a similar manner 24 hours after treatment.

All eyes were examined 1, 24, 48, and 72 hours after instillation of the test substance. Examinations were also conducted 7 days after treatment. Ocular reactions were scored according to the following scales:

Corneal opacity

Degree of density

1 - scattered or diffuse area, details of iris visible
2 - easily discernible translucent areas, details of iris slightly obscured
3 - opalescent areas, no details of iris visible, size of pupil barely discernible
4 - opaque, iris invisible

Area of cornea involved

1 - one-quarter (or less but not zero)
2 - greater than one-quarter to less than one-half
3 - greater than one-half to less than three-quarters
4 - greater than three-quarters

score = score for degree x score for extent x 5
maximum = 80

Iris

1 - folds above normal, congestion, swelling, circumcorneal injection (any one or a combination of these), iris [illegible] reacting to

8. MATERIALS AND METHODS (continued)

light (sluggish reaction is positive)
2 - no reaction to light, hemorrhage, gross destruction (any one or all of these)

score = score for iris x 5
maximum score = 10

Conjunctivae

Redness

1 - vessels definitely injected above normal
2 - more diffuse, deeper crimson red, individual vessels not discernible
3 - diffuse beefy red

Chemosis

1 - any swelling above normal (including nictitation membrane
2 - obvious swelling with partial eversion of the lids
3 - swelling of lids about half closed
4 - swelling of lids about half to completely closed

Discharge

1 - any amount different from normal (does not include small amount in inner canthus of normal animals)
2 - discharge with moistening of the lids and hairs just adjacent to the lids
3 - discharge with moistening of the lids and considerable area around the eye

Score = sum of values for redness, chemosis, and discharge multiplied by 2. Maximum = 20

The test substance was classified according to the following categories:

I. Corrosive (irreversible destruction of occular tissue) or corneal involvement or irritation persisting for more than 21 days.

II. Corneal involvement or irritation clearing in 8-21 days.

8. MATERIALS AND METHODS (continued)

III. Corneal involvement or irritation clearing in 7 days or less.

IV. Minimal effects clearing in less than 24 hours.

9. REPORTED RESULTS

The reported mean corneal and iris scores were 0.1 at 24 hours, and at all other examinations those scores were 0.

Mean scores for the conjunctivae were 1.1, 0.8, 0.8, 0.4, and 0.0 for redness at 1, 24, 48, and 72 hours and 7 days, respectively; respective scores for chemosis were 0.1, and 0.4 for the 1 and 24 hour observations and 0 at all other examination times.

10. DISCUSSION

The report included adequate information to support the conclusions of the investigators.

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DATA EVALUATION RECORD

1. CHEMICAL: Terbutryn
2-tert-butylamino-4-ethylamino-6-methylthio-s-triazine

2. TEST MATERIAL: Igran® 80 WDG (76% active ingrediant, 4% related compounds)

3. STUDY/ACTION TYPE: Skin sensitization - guinea pig; ("Me too" registration)

4. STUDY IDENTIFICATION: Maedgen, J. L., E. J. Sabol, R. J. Sabol, R. Mendez, and L. D. Weidner. June 28, 1984. Guinea pig sensitization: Igran® 80 WDG FL 840804. Unpublished report prepared by Stillmeadow, Inc., Houston, TX. Submitted by Ciba-Geigy Corp., Agriculture Division, Greensboro, NC. EPA Acc. No. 255226.

5. REVIEWED BY:

Name: Roger Gardner
Title: Toxicologist
Organization: Review Section 6
Toxicology Branch
Signature: Roger Gardner
Date: 4-1-85

6. APPROVED BY:

Name: Jane Harris, Ph. D.
Title: Section Head
Organization: Review Section 6
Toxicology Branch
Signature: Jane Harris
Date: 4/1/85

7. CONCLUSIONS: The results of the study indicate that Igran 80 WDG should not be classified as a skin sensitizer in guinea pigs.

Core classification: Minimum

8. MATERIALS AND METHODS

Test species: Male Hartley albino guinea pigs weighing from 315 to 380 g were used.

Experimental procedure: The hair was clipped from the backs of test animals 24 hours prior to the initial and final treatments (see below). Similar preparations were made on the second through the 21st days of the study.

Two groups of 10 animals were used. The first group received

an initial application of a 0.05% solution of dinitrochloro-

8. MATERIALS AND METHODS (continued)

benzene (DNCB) in ethanol, while the second group received an application of a 10.0% solution of the test substance in deionized water. The report noted that the 10% concentration was selected on the basis of a preliminary study and is considered to be the highest non-irritating concentration that can be used. No further discussion of the preliminary study was included in the report.

Applications were made under 5/8 by 9/8 inch gauze pads secured to prepared skin sites by a 1.5 by 2 inch piece of adhesive tape. The animals were treated on days 1, 3, 6, 8, 10, 13, 15, 17, 20, 22, and 36 of the study. Body weights of the test animals were obtained on days 0, 7, 14, 21, 28, and 35. Exposures lasted for 6 hours, and at the end of that time, the gauze pads were removed and test sites were examined and scored for reactions.

Erythema and eschar formation as well as edema were scored on a 5-point scale (0-4) with a maximum possible score of 8 for any site. Scoring was done according to the following classifications:

Erythema and eschar		Edema	
No erythema	0	No edema	0
Slight erythema	1	Very slight edema	1
Well-defined erythema	2	Slight edema	2
Moderate to severe erythema	3	Moderate edema	3
Severe erythema to slight eschar formation	4	Severe edema	4

Each test site was scored 24 hours after application of the test substance. In addition, sites were scored 48 hours after the applications which were made on days 1, 10, and 36. The authors stated that an average irritation score was obtained by adding scores for each time period and dividing by the number of observations for that time period. The authors further noted that a sensitizing reaction was indicated by an increase in positive reactions after the final dose (on day 36) in comparison to the reactions after the first dose (on day 1).

9. REPORTED RESULTS

The average irritation score after the first dose in the positive control group was reported to be 0.0, and the reported average after the last DNCB dose was 2.2. The two reported

9. REPORTED RESULTS (continued)

average irritation scores for those animals treated with the test substance were 0.0 after the first and final treatments.

10. DISCUSSION

Adequate information was included in the report to support the conclusions of the authors.

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DER
Addendum to Review
of Two Year Chronic Rat Toxicity Study

Chemical: Terbutryn

Study Title: Two-Year Chronic Oral Toxicity Study in Rats (IRDC Report No. 382-008; March 27, 1980)

Prepared By: Judith W. Hauswirth, Ph.D. Judith W Hauswirth 8/25/80
Mission Support Staff
Toxicology Branch/HED

Approved By: Reto Engler, Ph.D., Chief
Mission Support Staff
Toxicology Branch/HED

Originally Reviewed By: William Dykstra, Ph.D.

Date of Original Review: July 25, 1980

This study was originally classified as Core-Minimum data. Terbutryn was found to be oncogenic at the highest dose tested (3000 ppm); however, an NOEL for systemic toxicity was not determined.

This study was rereviewed in preparation of the registration standard for terbutryn. The NOEL for chronic toxicity in the rat is difficult to determine since complete hematologic and biochemical analyses were not done at all time points for all dosage groups. Hematology was done at 3, 6, 12, 18 and 24 months on the control and high dose groups (10 rats/sex/group) and at 12 and 18 months for the low and mid dose groups. Erythrocyte and hemoglobin values were significantly decreased at 18 months for all dosage groups. Twenty-four month values were not available for the low and mid dose groups but were significantly reduced in the high dose group. Biochemistry was done at 3, 6, 12, 18 and 24 months for the control and high dose groups (10 rats/sex/group) and on 5 rats/sex at 12 months only for the low and mid dose groups. SGOT values were significantly elevated at the 3 and 6 month sampling periods and alkaline phosphatase at 12, 18 and 24 months in the high dose groups.

Because of the deficiencies noted above this study is reclassified as Core-Supplementary. To upgrade this study to Core-Minimum a special 24 month study in male and female rats of this strain will be necessary to determine an NOEL for the effects of terbutryn on hematology and clinical chemistry. Histopathology and urinalysis need not be a part of this study. The dosages should carefully chosen. The fact that other effects due to terbutryn were seen at 3000 ppm (decreased body weight gain) should be considered. Ten rats/sex/group should be used. Blood should be taken for analysis at 3 month intervals.

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

JUL 25 1980

MEMORANDUM

OFFICE OF TOXIC SUBSTANCES

SUBJECT: EPA Reg.#100-540, Terbutryn; 6(a)(2) data
CASWELL#1250, Accession#242568-73

FROM: William Dykstra
Toxicology Branch, HED (TS-769) WHD 6/26/80
CT 7/1/80

TO: Robert Taylor (25)
Registration Division (TS-767)

Recommendations:

1. The memo of January 20, 1978 from John Doherty to Robert Taylor indicates that some studies submitted were performed by I.R.T. and are required to be validated. These studies are I.R.T.#8904, A5458, A5457.

2. The 6-month dog study is acceptable as core-minimum data. The NOEL is 10 mg/kg/day (400 ppm).

3. The two-year mouse oncogenicity study is acceptable as core-minimum data. No oncogenic effects resulted from terbutryn at dietary levels up to 3000 ppm.

4. The two-year rat chronic toxicity oncogenicity study is acceptable as core-minimum data. Statistical analysis of the tumor incidence in the high-dose group, using Chi-Square one-sided test with Bonferroni adjustment, showed statistically significant incidences of thyroid follicular adenomas ($p < .05$), female mammary gland adenomas ($p < .05$) and female liver adenomas ($p < .05$). Male testicular cell adenoma ($p = 0.0752$) and female liver focal cytomegaly ($p = 0.08568$) were of borderline significance.

   These oncogenic effects from terbutryn fed rats trigger an oncogenic RPAR criteria.

5. The 3-generation reproduction study is acceptable as core-minimum data. The NOEL for the study is 300 ppm.

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Review:

1) Six-Month Oral Toxicity Study of Terbutryn Technical in Beagle Dogs (Elars Bioresearch Laboratories, Project No. 1421, Jan. 22, 1980)

Test Material: terbutryn technical, ARS#2080/78, Batch#FL-761552

Sixty purebred Beagle dogs, equally divided by sex and approximately four months of age, were used in this study. The dogs were identified by ear tatoos and individually housed in stainless steel cages in an environmentally controlled room. High Protein was fed free choice with feed consumption measured daily for a weekly total. Fresh tap water was provided ad libitum. The dogs were dosed orally once a day for at least 180 days consecutively.

Treatment Design

Group	Daily Dose (mg/kg)	Number of Dogs
Control	Control - Sham dosed	8M, 8F
T-I	10 mg/kg	6M, 6F
T-II	25 mg/kg	6M, 6F
T-III	50 mg/kg	8M, 8F

Observations were made daily from seven days predose to the day before the individual dog was necropsied. Each dog had a separate sheet which contained parameters such as appecite, elimination, appearance, behavior and accompanying comments. Food consumption and body weight were measured weekly.

Opnthalmologic examinations were done at predose and at termination.

Specimens for clinical pathology were collected at 10 days pre-dosing and every month for the six month duration of the study. Hematology parameters included: leukocyte (total and differential), erythrocyte and platelet count, hematocrit, hemoglobin, mean corpuscular volume (MCV), retriculocyte count, methemoglobin, Heinz bodies, protime and activated partial thromboplastin time (APTT).

Clinical chemistry determinations included: glucose, BUN, total protein, total bilirubin, direct bilirubin, cholesterol, calcium, potassium, SAP, SGOT, SGPT and LDH.

Urine collection for urinalysis was done 8 days prior to initiation of dosing and then at 2, 4 and 6 months into the study. Urinalysis determinations included: color, specific gravity, pH, protein, glucose, ketones, bilirubin and urobilinogen, nitrite, blood, leukocyte count, erythrocyte count and the presence of epithelium, bacteria and triple phosphate crystals.

For the above tests, the dogs were fasted for 12 hours.

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Dogs (AT08) of T-I was necropsied on March 11, 1979, day 2[illegible] of the study.

All dogs were fasted for at least 12 hours before sacrifice.

Gross observations were performed and samples of the organs listed below were taken.

Adrenals (2)
Esophagus
Heart
Colon
lung with bronchi
mammary gland (female)
muscle
trachea
ovaries or testes
pituitary
mandibular salivary gland (2)
parotid salivary gland (2)
small intestine
spleen
thymus
tissue mass (if present)
urinary bladder - ureters
Any gross lesions (if present)
Aorta
Brain (pons, cerebrum, cerebellum)
Epididymes (2)
gall bladder
Kidneys (2)
Liver
lymph nodes
nerve
pancreas
prostate (male)
skin
spinal cord
stomach
thyroids & parathyroids (2)
uterus
Bone marrow (sternum)
eyes (with optic nerve)

The following organs were trimmed in a uniform manner and weighed: heart, spleen, liver, kidneys (2), adrenals (2), thyroid and parathyroid (2), brain, testes (2), or ovaries (2) and pituitary.

Eight dogs, two males and females from control and T-III were allowed to undergo a recovery period of 29 days. These dogs were treated in exactly the same manner as during the six month study, with the exception that they were no longer given test material.

Results

One dog in TA-II (KR08) and five dogs in T-III (GD08, QW08, HF08, AD08 and FU08) showed symptoms which could be compound related. These dogs exhibited increased salivation, a pronounced response to sharp noises, grinding of teeth, and a marked change in behavior with the dog becoming increasingly apprehensive. There appeared to be no set trend or pattern concerning onset of symptoms which could occur irregularly during the six month period and last from four to six hours.

The compound seemed to have a more pronounced effect on one of the T-III dogs (GD08) than on any of the other dogs. On Feb. 21, 1979 and June 15, 1979, the dog exhibited a mild proprioceptive deficit manifested by an inability to rest on both knuckled hind feet to a normal position. The placing reflex was slow for both hind legs and the dog appeared dull, apprehensive, and unresponsive.

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In conclusion, the dog manifested a mild to moderate posterior ataxia which lasted for approximately 24 hours. There was no significant differences in food consumption between the control and treatment groups. However, there appears to be a trend of reduced weight gain with increasing dose for the two higher levels. There were no lesions consistent through any dose group and no dose-related lesions observed in the ophthalmoscopic examination.

There were no significant differences in urine analysis between the control and treatment groups during the time period being considered. In addition, no trends could be detected.

In hematology, there appears to be a trend in the platelets count although there is not a significant difference. All groups of males show a decrease from predose to six months (termination) with T-III showing the greatest decrease. For females the trend is not as prominent. Significant differences from control values occur for T-I males at three months for reticulocytes and five months for APTT. For T-II males the significant difference occurs at six months for monocytes and T-II females at four months for hemoglobin.

These values, however, are within normal limits for these parameters and the values did not continue to be significantly different from control during succeeding bleeding periods. Significant differences were detected in cholesterol values of T-II females for months one and two, in the calcium values of T-III males for month two, and in direct bilirubin of T-I females for month three. Most of the values that were found to be significantly different were within the normal limits.

No drug related trend was found in electrophoretic scans of total protein. With respect to organ weights, there appears to be no significant difference between the control and treatment groups.

A T-I male dog in a moribund condition, AT08, was necropsied on March 11, 1979, the 24th day of the study. On gross examination, the lungs were dark red to pink in color, firm, congested and edematus. A forthy fluid was visible in the trachea. The mediastinal lymph nodes were swollen and edematous. A firm mass (9 x 13 mm) was attached to the gall bladder.

At necropsy three out of twelve nonrecovery dogs in the T-III group (50 mg/kg) had evidence of edema hemorrhage or mucosal thickening of various segments of the small intestine. Microscopic examination of these areas revealed evidence of hyperemia, and hemorrhage of Peyer's patches, intestinal mucosa and intestinal musculature.

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The necropsy and microscopic examination of the four recovery dogs that were in Group T-III did not reveal any evidence of small intestine mucosal or musculature alterations. One dog in the Group III recovery animals (0408) did have gross and microscopic signs of very slight lymphoid hyperplasia in the colon. At necropsy one out of twelve animals in the T-II group (25 mg/kg) had evidence of thickening of the mucosa in segmental areas of the small intestine. Microscopic examination revealed very slight evidence of hyperemia in the Peyer's patches of the ileum, but the remainder of the small intestine samples were judged not remarkable. In the T-I group of animals, one animal was noted at necropsy to have a reddened area of mucosa in the jejunum. At microscopic examination this tissue sample was diagnosed as very slight mucosal congestion. It was also noted that this animal did not exsanguinate well at terminal sacrifice, which could account for the mucosal congestion.

Conclusion:

The low-dose dogs (10 mg/kg/day) in Group T-I are considered the NOEL for the study. The LEL is 25 mg/kg/day (T-II).

Classification: Core-Minimum Data.

2) Two-Year Carcinogenicity Study in Mice (IRDC Report#382-005; April 21, 1980)

Test Material: terbutryn technical; ARS No. 2046/76; Batch No. FL-761552, 64 lbs.; white powder.

Two-hundred forty male and 240 female Charles River CD-1 mice, weighing from 23 to 32 grams and 18 to 24 grams, males and females respectively, were initiated in this study. The mice were housed individually in suspended wire-mesh cages and maintained in a temperature-, humidity- and light-controlled room. Water and the control and test diets were available ad libitum.

The mice were ear punched to identify the dosage-level groups. Ear punches were verified at each cage change and before necropsy. Terbutryn technical was offered daily in the diet at dosage levels of 3, 1000, and 3000 ppm. Sixty male and 60 female mice were used at each dosage level and in a control group. The control group received basal laboratory diet only, on the same regimens as treated mice.

The mice were observed three times daily (Monday-Friday) or twice daily (weekends and holidays) for signs of overt toxicity, moribundity and mortality. Detailed observations were recorded weekly. Individual body weights and sex-group food consumptions were recorded every four weeks.

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All surviving mice were sacrificed and necropsied at termination of the study. Necropsy examination consisted of examination of the external body surface and body orifices. The mouse was then opened and the contents of the body cavities were examined in situ, removed and again examined. All organs, tissues and remaining eviscerated carcass were then collected and mixed in phosphate buffered 10% formalin. Mice which died or were sacrificed in extremis during the course of the study were necropsied as above.

Hematoxylin and eosin stained paraffin sections of the following tissues were prepared by standard histologic methods and examined microscopically from all mice from the control group and the 3000 ppm dietary group. Microscopic examination of tissues from the 3 and 1000 ppm dietary groups was limited to gross lesions observed at necropsy.

Brain	thyroid
spinal cord (3 sections)	parathyroids
sciatic nerve	adrenals
eye	trachea
optis nerve	lung
pituitary	heart
spleen	aorta
liver	stomach
kidney	small intestine
urinary bladder	large intestine
testes or ovaries	pancreas
prostate or uterus	lymph nodes
mammary gland (females)	bone marrow (sternum)
salivary gland	skeletal muscle
esophagus	any other tissue with
skin	gross lesions

Results

No changes considered to be related to compound were observed in general behavior and appearance. No difference considered to be related to compound were observed in body weight changes of treated mice as compared with controls. No differences considered to be related to compound were observed in the food consumption values of treated mice as compared with controls.

Survival at week 104 was as follows:

Dosage Level ppm	Number Surviving/ Number Initiated	
	Male	Female
0 (control)	29/60	35/60
3	29/60	34/60
1000	28/59*	30/60
3000	34/60	38/60

*one mouse missing

No gross pathologic lesions which were considered related to Terbutryn technical were observed in any mice from the experimental groups. No microscopic pathologic lesions which were attributable to Terbutryn technical feeding were observed in any tissues examined from any mice from the experimental groups.

Conclusion:

Terbutryn technical was not oncogenic to mice at dietary levels up to 3000 ppm.

Classification: Core-Minimum Data

3) 2-Year Chronic Oral Toxicity Study in Rats (IRDC Report No. 392-008; March 27, 1980)

Test Material: Terbutryn technical, ARS No. 2046/76, Batch No. FL-761552, 65 lbs.

Two-hundred sixty male (weighing from 128 to 225 gms) and 260 female (weighing from 115 to 181 gms) weanling Charles River CD rats were initiated in this chronic oral toxicity study. Five animals of each sex from the control and 3000 ppm groups were sacrificed and necropsied after 12 months of study. At that same time, a second 5 rats/sex in each of these groups were placed on a control diet for 4 weeks before being sacrificed and necropsied. The rats were housed individually in hanging wire-mesh cages and maintained in a temperature-, humidity- and light-controlled room. Metal ear tags were placed on the rats approximately 3 months after study initiation.

The rats were randomly distributed into groups and offered the appropriate diets as follows:

Dosage Level ppm	No. of Rats/Group Male	Female
0 (control)	70	70
2	60	60
300	60	60
3000	70	70

The rats were observed twice daily for signs of overt toxicity and mortality. Detailed observations were recorded weekly. Individual body weights and food consumption (10 rats/sex/group) were recorded weekly for the first 3 months and monthly thereafter.

During the 4-week withdrawal period, individual body weights and food consumption were determined weekly for the recovery rats. At 3, 6, 12, 18 and 24 months, blood for hematologic and biochemical studies and urine for urinalysis were collected from 10 rats/sex from the control and high-dose groups. At 12 and 18 months of study, rats from the

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low- and mid-dose groups were also selected for testing. Hematologic examinations were done for the recovery rats after they had completed 4 weeks of compound withdrawal.

Microscopic examinations were conducting upon the following tissues, preserved in buffered neutral 10% formalin, taken from the 12-month interim sacrifice, all surviving animals in the control and high-dose groups at the 24 month sacrifice, as well as from all rats in these groups which died on study. These were prepared by standard histologic method using hematoxylin and eosin.

stained parafin sections	pancreas	aorta
adrenal gland	bone marrow	sciatic nerve
parathyroid	pituitary	cecum
brain	colon	skin
salivary gland	small intestine	eye
esophagus	spinal cord	harderian gland
gonads	heart	sternum
spleen	stomach	liver
kidney	thyroid	lymph node
lung	urinary bladder	mammary gland
trachea	muscle	optic nerve
uterus		

The following tissues were taken from all low- and mid-dose animals at 24-month sacrifice:

testes	thyroid
liver	mammary gland

Significant gross lesions were also examined microscopically. All tissues from the 12-month sacrifice are rats which died on study up until 12-months were processed and examined microscopically at IRDC. Tissues from all other animals were processed and examined by Experimental Pathology Laboratories (Dr. J.F. Ferrell, D.V.M.). Statistical analysis of the data was performed.

Results

There were no signs of overt toxicity noted for the treated rats in the regular and recovery groups. No compound-related effects were observed on the survival rate of the treated rats.

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Survival at 104 weeks of study was as shown below:

Dosage Level (ppm)	No. Survivors/No. Initiated	
	Male	Female
0 (control)	38/60	35/60
2	41/60	32/60
300	47/60	40/60
3000	44/60	38/60

Survival for the 4-week recovery period was 100%.

The high-dose males and females showed a large decrease in body weight throughout the study [week 104: males (-21.7%) and females (-39.0%)]. Decreases were noted in the food consumption values over the 2-year study for both male and female rats at the high-dosage level. No changes indicative of a compound effect were noted in the hematologic profile or urinalysis of the groups tested.

Blood biochemical values showed some apparent compound-related effects in the high-dose females.

Significant changes were seen in the fasting blood sugar, SAP, SGOT and cholesterol values at various intervals. These effects indicate that the test material caused physiological significant changes.

There were occasional statistically significant variations in organ weight but the biological significance of the variations noted in the organ weights is unknown.

The histopathological evaluation of non-neoplastic lesions indicated that, with the exception of focal cytomegaly, non-neoplastic lesions were randomly distributed between the control and 3000 ppm groups.

The focal pneumonitis was considered to be of dubious significance.

Statistical analyses of the tumor incidence demonstrated significant tumor incidences in the high-dose group. Thyroid follicular adenoma in males was statistically significant.

In females, mammary gland adenoma and liver adenoma were statistically significant. Borderline statistical significance occurred in testicular interstitial cell adenoma in males and liver focal cytomegaly in females. These results are summarized in Table I.

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Tumor Type	Control	Low Dose	Mid Dose	High Dose	Chi Square	P of 2 Side test	P of 1 Side test	With
Males								62
Thyroid follicular adenoma	1 65	0 59	0 60	7 65	4.795	.02930	.01465	
Males								
Testicular Interstitial cell adenoma	13 65	11 60	14 60	23 65	3.841	.05001	.02501	
Females								
Mammary gland adenomas	6 64	9 57	8 58	15 64	4.628	.03145	.01573	
Females								
Liver focal cytomegaly	9 64	13 59	9 59	18 65	3.619	.05712	.02856	
Females								
Liver adenoma	3 64	2 59	3 59	12 65	5.954	.01468	.00734	

* = significant
$p < .05$
$\underline{8}$ = borderline significant

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Conclusion

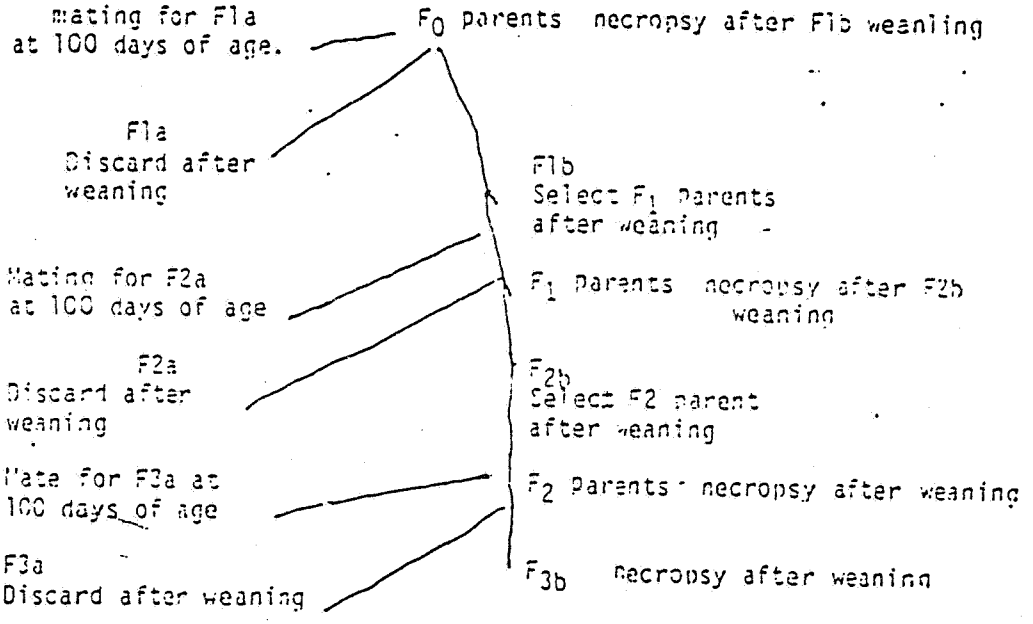
Terbutryn technical was oncogenic at the high-dose in both males and females.

Classification: Core-Minimum Data

4) Three-Generation Reproduction Study in Rats (IRDC Report No. 382-011, Oct. 8, 1979)

Test Material: terbutryn technical, ABS No. 2046/76, Batch No. FL-761552, 55 lbs., white powder

In this three generation reproduction study in Charles River-CD rats, the test material was administered in the diet at dosage levels of 0, 300 and 3000 ppm. Forty male and 80 female rats were evenly distributed among the three treatment groups and one control group. Each generation was mated twice to produce two litters. The matings scheme is shown below:



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Results

No changes considered to be related to treatment of test material were seen in relation to the general behavior, appearance or survival of the treated parental rats when compared to the controls. Dam#41958AA in the F_2 generation (3000 ppm) died and was replaced by dam#419748C on study week 65.

Moderately to severely reduced mean body weights were seen in the 3000 ppm groups during the entire generation of the F_0 parents, with significant differences mean body weights reported at study weeks 9 and 34 when compared to the control groups. In the F_1 generation at the 3000 ppm level, severely reduced mean body weights in the males and moderately to severely reduced mean body weights in the females were seen as compared to control values with significant differences in mean body weights at the $p < .01$ level at all points of analysis (weeks 33, 42 and 64).

In the F_2 males, slightly to moderately reduced body weights were seen throughout the entire generation in the 2 ppm and 300 ppm dosage groups; but, there were no statistically significant differences in these changes as compared to control values. Severely reduced mean body weights were observed in the 3000 ppm group, with a significant difference in mean body weight at the $p < 0.01$ level when compared to the control group at all points of the analysis (weeks 63, 73 and 94).

At the 3000 ppm level, moderate to severe decreases in mean body weight were also seen in the F_2 females during the entire generation, with significant differences in mean body weights at the $p < 0.01$ level when compared to the control groups at all points of analysis (weeks 63, 73 and 94). All other treatment groups were comparable to the control groups in respect to parental body weights.

Based on the average of mean food consumption values of parental rats, a slight decrease in mean daily food consumption was observed in the F_0 males in the 3000 ppm dosage group as compared to controls.

The F_0 females exhibited a slight decrease in mean daily foood consumption values in the 300 ppm group and a moderate decrease in the 3000 ppm group, as compared to the control group. All other treatment groups showed no meaningful or biological differences when compared to the control group in mean daily food consumption.

Lower female fertility indices were seen for the F_0 females at the 3000 ppm dosage level in both the F1a and F1b litters, with a statistically significant difference at the $p < .05$ level reported in this high dose group in the F1b litter when compared to the control group.

In the F3a litters, a lower male fertility index was observed at the 3000 ppm level; but there was no statistically significant decrease observed in the 5 day survival index at the 3000 ppm level ($p < 0.01$) in

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the F1a litters when compared to their respective control. A statistically significant decrease was also observed in the number of live pups at day 0 of lactation at the 300 ppm level in the F1a litters when compared to control values. At the 2 ppm level in the F1b litters, a statistically significant increase was reported in the number of live pups at day 0. However, this difference was attributed to random occurrence and was not considered a result of treatment.

No other differences were observed with respect to male and female fertility, the length of the gestation periods and the viability and survival of the pups through weaning.

The mean pup body weights at lactation day 21 of each of the six litters produced (F1a, F1b, F2a, F2b, F3a, F3b) at the 3000 ppm treatment level were statistically significantly lower ($p < 0.01$) than the mean weights of the control pups. Also, the mean pup weights of both sexes in the F2a at the 300 ppm treatment level were statistically lower ($p < 0.05$) when compared to the control values. No significant gross lesions were observed in the F_0 and F_1 parents. In the F_2 parental females, two animals each of group II and III and three animals of group IV out of 10 females in each group had hydrometra. In the F3b generation weanlings, two males and three females of group IV were smaller in size. The animals dying on study showed mainly autolysis. One animal in the 2 ppm F2 parental group had protrutary enlargement.

No histopathological change of significance was observed in F_0 and F_1 parents three out of 10 animals in group IV had atrophy of the endometrium with dilatation of the uterine lumen to a moderate degree. In the F3b generation weanlings belonging to group IV, two males and two females of 10 of each sex had very slight to slight lymphoid hyperplasia in the colon.

Statistically significant variations in the absolute/relative weights of organs of the parental rats were seen as follows:

F_0 Parents

Organ	Dosage Level (ppm)	Sex	Weight	Change	p<
liver	3000	M	relative	increase	0.05
	3000	F	relative	increase	0.01
kidneys	3000	F	relative	increase	0.01
gonads	3000	M	relative	increase	0.05
heart	3000	M	absolute	increase	0.01
	3000	M	relative	increase	0.01
	3000	F	relative	increase	0.05
brain	3000	M	relative	increase	0.01
	3000	F	relative	increase	0.01

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F_1 Parents

Organ	Dosage Level (ppm)	Sex	Weight	Change	p<
liver	300	M	absolute/relative	increase	0.05,0.01
	3000	M	relative	increase	0.01
	3000	F	relative	increase	0.01
kidneys	2	M	absolute	increase	0.05
	300	M	absolute	increase	0.01,0.05
	3000	M	relative/relative	increase	0.01
	3000	F	absolute	decrease	0.05
spleen	3000	F	absolute	decrease	0.05
gonads	2	F	absolute/relative	increase	0.05,0.01
	3000	M	relative	increase	0.01
brain	3000	M	relative	increase	0.05

F_2 Parents

Organ	Dosage Level (ppm)	Sex	Weight	Change	p<
liver	2	M	absolute/relative	decrease	0.05
	3000	M	absolute	decrease	0.01
	3000	F	absolute	decrease	0.01
kidneys	3000	M	absolute/relative	decrease	0.05
	3000	F	absolute/relative	decrease	0.05
gonads	3000	M	relative	increase	0.01
	3000	F	relative	increase	0.05
heart	3000	M	absolute/relative	decrease/ increase	0.01
	3000	F	absolute	decrease	0.01
brain	3000	M	relative	increase	0.01
	3000	F	absolute/relative	decrease/ increase	0.05

Conclusion

The NOEL for the study is considered to be 300 ppm. Although, effects on number born alive occurred in F1a and a significant decrease in pup body weight occurred in F2a, these effects did not occur in any other several at this dose level and are therefore not considered biologically meaningful. The significance of the change in organ weights is unknown since no histopathology was observed. The lowest effect level (LEL) is 3000 ppm (the high dose group).

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Classification: Core-Minimum Data

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DER
Addendum to Original DER
for Teratology Studies on Terbutryn

Chemical: Terbutryn

Prepared By: Judith W. Hauswirth, Ph.D. Judith W. Hauswirth 6/26/86
Mission Support Staff
Toxicology Branch/HED

Approved By: Reto Engler, Ph.D., Chief
Mission Support Staff
Toxicology Branch/HED

Originally Reviewed By: Alex Arce

Original Date of Review: January 1986

Background

Two teratology studies on terbutryn, one in the rat, the other in rabbits, were initially reviewed in January 1986 by Toxicology Branch (see attachment). These studies were rereviewed in May 1986 for the Registration Standard on terbutryn. Upon rereview the NOEL for developmental toxicity in the rabbit was changed from 75 to 50 mg/kg and the NOEL for maternal toxicity in the rat was changed from 10 to 50 mg/kg. The reasons for these changes, as well as other points, not brought out in the original review are discussed herein.

Review

1. A Teratology Study in New Zealand White Rabbits. Ciba-Geigy Pharmaceutical Division, Reproductive and Genetic Toxicology Subdivision. October 1, 1985.

o Maternal Toxicity

In the original review the following was stated:

"Food consumption: Decrease at the high-dose level during gestation period. The other groups also showed decreases. Such observations were significant even at the lowest treatment dose.

Body weight: Although no significant differences between treated and control mothers were observed, the variations in maternal body weights were significant at various intervals and they were related to the variations in feed consumption."

The Food Index was calculated for each dosage group at various time periods to determine the significance, if any, of the decreased food consumption apparently seen at all dosage levels of terbutryn. Rabbits were dosed by gavage from days 7-19 of gestation.

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Food Index Calculation*
Group

	0	10	50	75
Days 0-7				
g. gained	232	205	136	215
food consumption(g)	1654	1575	[illegible]	1642
Food Index	7.9	7.6	[illegible]	7.6
	0	10		75
Days 7-14				
g. gained	117	77	43	3
food consumption(g)	1529	1385	1305	1005
Food Index	12	18	30	335
Days 14-19				
g. gained	122	43	89	-4
food consumption(g)	1127	882	909	546
Food Index	9	20	10	-136

*Food Index (F.I.) = $\frac{\text{grams food consumed}}{\text{grams b. wt. gain}}$

At days 7-14 the F.I. is considered by this reviewer to be significantly elevated in the 50 and 75 mg/kg groups, and at day 14-19 in the 75 mg/kg group only. The increase in F.I. at 10 mg/kg (days 14-19) was not considered treatment related since it was not seen in the 50 mg/kg group which was comparable to the untreated control group.

An NOEL was determined for maternal toxicity. Based upon changes in food consumption, F.I., body weights, and stools at 50 mg/kg the NOEL for maternal toxicity is 10 mg/kg in the rabbit.

o The original review states that there were no treatment related changes in developmental toxicity related to terbutryn treatment. This reviewer disagrees with this conclusion and believes that the NOEL for developmental toxicity is 50 mg/kg based upon reduced ossification of sternebrae in rabbits at 75 mg/kg.

Incidence of Reduced
Ossification of Sternebrae

Group (mg/kg)

	0	10	50	75
By fetus	14	31*	39*	39*
# examined	129	152	164	123
% affected	10.9	20	24	31.7

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By litter	8	14*	14*	12*
# litters	15	18	19	14
% affected	54.4	77	73.6	85.7

Historical Control Information (14 Studies)
By fetus: mean = 14.98%
range = 1.4 - 29.0%
By litter: mean = 50.23%
range = 11.1 - 73.3%

* $p<0.05$

Although the increased incidence of reduced sternebrae ossification was significant at all dosage levels, it was outside of the historical control range when expressed by fetus or litter only at 75 mg/kg. The increase seen at 75 mg/kg is considered by this reviewer to be related to terbutryn treatment. Therefore, the NOEL for developmental toxicity for the rabbit is 50 mg/kg. The Core-Classification remains the same, Minimum.

2. Teratology Study in Rats. Ciba-Geigy Corporation, Research Department and Genetic Toxicology Subdivision. October 25, 1985.

This reviewer disagrees with the NOEL determined for maternal toxicity in the original review of this study. The review states that the NOEL of 10 mg/kg was based upon mortality, weight loss, salivation, urine staining and blood discharge. However, all of these effects, except salivation were seen at the high dose (500 mg/kg) and not at the mid dose (50 mg/kg). The incidence of salivation was 0/25 (control), 0/25 (low dose), 4/25 (mid dose) and 22/25 (high dose). At the mid dose, salivation was seen prior to dosing in three of the animals and only on one day for each animal post-dosing. This reviewer does not feel that this finding is toxicologically significant nor treatment-related at the 50 mg/kg dose level.

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DATA EVALUATION REPORT

Chemical: Terbutryn.

Test Material: Terbutryn Technical

Study Type: Teratology Report No. 85010

Title: A Teratology Study in New Zealand White Rabbits (MIN 842105)

Laboratory: Ciba-Geigy Pharamaceutical Division, Reproductive and Genetic Toxicology Subdivision

Location: Summit, NJ 07901 Date: October 1, 1985

Lab. No.: Ref. No. I-023-02

Sponsor: Agricultural Division of Ciba-Geigy

EPA ID No.: 080813 MRID: None Accession No.: 289886

Reviewed by: Alex Arce
Toxicologist
Toxicology Branch
Hazard Evaluation Division (TS-769C)
Date: January 1986
Phone: 557-7457

Approved by: Clint Skinner, Ph.D.
Section III Head
Toxicology Branch
Hazard Evaluation Division (TS-769C)
Date:
Phone:

Conclusion:

Developmental Toxicity

Developmental NOEL = 75 mg/kg (HDT)

A/D ratio 10/75 = 0.13

-Maternal NOEL = 10 mg/kg LEL = 50 mk/kg

Levels tested by gavage in New Zealand White Strain ; 0,10,50 and 75 mg/kg /day
Core Classification: Guideline

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Protocol:

Test Material and Methods:

Material: Terbutryn Technical
3% corn starch containing 0.5% Tween 80

Animals: New Zealand White Rabbits

Sex: female; Age: adult; Weight: Acceptable;
Source: Not established.

Dosage: 0, 10, 50 and 75 mg/kg/day
Number of animals per dose level: 19 mothers/dose level

Description:

The product was administered by gavage to pregnant rabbits during the 7th to 19th day of gestation in three dose levels, daily. The rabbits were necropsied on day 29 and mothers of fetuses were examined and weighed. Visceral abnormalities were recorded.

Reported Results: (Refer to attached addendum extrated from submitted data)

Deaths: None

The food consumption and body weight gain in all groups decreased. The product is reported to be not embryotoxic, fetotoxic or teratogenic. Induced loss weight in the mothers at all doses.

Observations:

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Signs of Toxicity: Daily

Behavior: Daily

Body Weights: On days 0, 7, 14, 19, 21, 25 and 29.

Food Consumption: Daily from day 0 to day 28.

Examinations:

Laparohysterectomy was performed, livers were weighed. ovaries were examined, corpora lutea counted, dead fetuses and absorptions were counted, the portions of the fetuses in the uterus were recorded. The fetuses were measured and weighed.

Maternal Examinations: (Extrated from Submitted protocol)

All mothers were examined for external and internal gross malformations or pathological changes. Representative samples of gross lesions were collected for microscopic observations.

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Fetal Examinations:

(From submitted Data)

Fetal Dissection:

On the day of necropsy (if possible) each fetus was examined viscerally according to a modification of the Staple technique (Staples, R.E., Teratology 9 (3): A-37, 1974) and its sex determined. The visceral examination was conducted on all fetuses as soon as possible following necropsy (not more than 24 hours). Visceral examination includes the following systems, organs and glands which were examined using dissection and slicing under appropriate magnification:

Central Nervous System:	brain
Cardiovascular System:	heart, major blood vessels
Respiratory System:	trachea, lungs, diaphragm
Gastrointestinal System:	oral cavity, tongue, esophagus, stomach, intestines, liver, gallbladder, pancreas
Lymphoid Structures:	thymus, spleen
Urinary System:	kidneys, ureters, bladder
Endocrine System:	adrenals
Genital System:	ovaries, uterus or testicles

Visceral examination data were collected on the standard R&GT visceral collection forms, the results of these data were tabulated manually (Appendix 19A), and the tabulations sent to the Statistics Section for analysis. Results of the visceral exams are recorded as normal or abnormal in the raw data; whereas, only abnormal data are presented and summarized in this report.

The fetuses were then prepared for a subsequent skeletal examination after clearing in potassium hydroxide and staining with Alizarin Red S (Staples and Schnell, Stain Technology 39: 62, 1964). This method is a modification of the method described by Dawson (Dawson, A.B., Stain Technical 1, 1926, p. 123-124).

Skeletal Examination:

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Following the visceral examination, all fetuses were stained and subjected to skeletal examination using appropriate magnification. All ossification centers that are characteristically present at day 29 of gestation in this strain of rabbit were examined for: presence/absence, size, shape, location and relationship to adjacent ossification centers. The Reproductive Skeletal and Visceral Program (R.S.V.P.) system an in-house

developed and validated database management system was employed to collect the data from the skeletal evaluations and the printouts generated are the raw data.

Results: (Refer to attached data , extrated from submitted study)

Mortality: None for the mothers (Some does were sacrificed due to conditions not related to treatment)

Signs of toxicity: Variations in the feces at the mid- and high-dose group. Other observations were not related to treatment.

Food consumption: Decrease at the high-dose level during gestation period. The other groups also showed decreases. Such observations were significant even at the lowest treatment dose.

Body weight: Although no significant differences between treated and control mothers were observed, the variations in maternal body weights were significant at various intervals and they were related to the variations in feed consumption.

Pathology: None of the various findings can be related to the administration of the compound.

Reproductive Observations:

No significant findings were observed.

Maternal Parameters:

Viable fetuses, number: normal.

Number of corpora lutes: no significant variations.

Sex of the fetuses: no significant variations.

Weights and Growth: no significant variations.

Number of litters: no significant variations.

Number of pregnant animals: no significant variations.

Number of abortions: not reported.

Fetal Parameters:

Length:

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Weight and Growth: no significant differences.

External observations:

Sex: no significant differences.

Visceral Observations: (Refer to attachment)

The various occurrences observed were noted at random in the three dose levels and the controls; thus they cannot be attributed to the administration of the material.

Skeletal Observations: (Refer to attachment)

The variations noted were regarded as general occurrences in rabbits; thus, not significant.

Alive fetuses: no significant differences. (Refer to attachment)

Dead fetuses: unremarkable.

Discussion:

The report is complete, the study was well designed and onducted. The conclusions are accurate and acceptable. The tudy is scientifically sound. The biological meaning of the ffects reported are that the product is not a teratogenic gent, it does induce developmental changes expressed as eduction in body weights but is not fetotoxic. The mechanism f these effects is related to the toxicity of the material hat affected the maternal body weights and food consumption, at andom. These effects mean that the material is safe for the ntended use, as per this test.

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DATA EVALUATION REPORT

Chemical: Terbutryn

Test Material: Terbutryn Technical

Study Type: Teratology

Title: Teratology Study in Rats (MIN 842292)
Report No. 85111

Laboratory: Ciba-Geigy Corporation, Research Department and Genetic Toxicology Subdivision

Location: Summit, NJ Date: October 25, 1985

Lab. No.: Not submitted.

Sponsor: Ciba-Geigy

EPA ID No.: 080813 MRID: None Accession No.: 289887

Reviewed by:

Alex Arce
Toxicologist
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Hazard Evaluation Division (TS-769C)
Date: January 1986
Phone: 557-7457

Approved by:

Clint Skinner, Ph.D.
Section III Head
Toxicology Branch
Hazard Evaluation Division (TS-769C)
Date: January 1986
Phone: 557-3710

Conclusion:

Core-Minimum Data (Requires Registrant's explanation)

Developmental Toxicity
NOEL = 50 mg/kg
LEL = 500 mg/kg
A/D ratio 10/50 = 0.2

Maternal toxicity NOEL = 10 mg/kg
LEL = 50 mg/kg
(Mortality, weight loss, salivation, urine stain, blood discharge)

Fetal toxicity NOEL = 50 mg/kg (HDT)
LEL = 500 mg/kg

Core Classification: Minimum

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were weighed and the fetuses examined for gross abnormalities, then placed into either 95 percent ethanol or Bouin's fixative for subsequent skeletal of visceral examination.

Fetal Examinations:

Approximately 1/3 of the fetuses were fixed in Bouin's solution for at least 1 week and then examined for visceral abnormalities according to the method of Monie, Kho, and Morgan (Supplement to Teratology Workshop Manual, pp. 163-169, 1965).

The visceral examination evaluates the following biological systems, organs and glands which are examined using dissection and slicing (where applicable) techniques under the appropriate magnification:

Central Nervous System:	brain
Cardiovascular System:	heart, and major blood vessels
Respiratory System:	nasal passages, trachea, lungs, diaphragm
Gastrointestinal System:	oral cavity, tongue, esophagus, stomach, intestines, liver, pancreas
Lymphoid Structures:	thymus, spleen
Urinary System:	kidneys, ureters, bladder
Endocrine System:	adrenals
Reproductive System:	ovaries, uterus or testicles

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Skeletal Examination:

Approximately 2/3 of the fetuses from each litter were stained with Alizarin Red S and cleared according to the method of Staples and Schnell (Stain Technology 39:62, 1964), and then examined for skeletal abnormalities.

The rodent skeletal examination involves checking, with the aid of appropriate magnification, all ossification centers that are characteristic of a rat fetus on gestational day 20 (Fritz, H. and Hess, R., Teratology, 3, 1972, Walker, D.G. and Wirtschafter, Z.T. The Genesis of the Rat Skeleton, A. Laboratory Atlas, Charles C. Thomas, Springfield, Illinois, 1967). The examination includes checking for the presence/absence, size, shape, location and relationship to adjacent ossification centers.

Results: (Refer to attached data extracted from submitted protocol)

Mortality: Mothers 2/25 found dead on day 20.

Signs of toxicity: Reduction of weight, feed consumption and weight gain, salivation, swollen abdomens, red stains around vulva, anus, face; lethargy and ptosis.

Behavior: Lethargic, at the high dose level

Body weight: Reduced at the high-dose level. Due to the large dose administered (500 mg/kg) such reported occurrences were expected.

Necropsy: Distended stomachs at the high dose level in most animals

Maternal Parameters:

Viable fetuses, number: significant (refer to conclusion).

Number of corpora lutes: no significant variations.

Sex of the fetuses: no significant effects on sex ratio.

Weights: severe at the high dose group.

Number of litters: no significant variations.

Number of pregnant animals: no significant variations.

Number of abortions: no significant variations.

Fetal Parameters: (Refer to attached data extrated from submitted protocol)

Length: reduced at the high-dose level.

Weight: decreased at the high-dose level.

External Observations: Pups

Weight decreased to 71 percent of controls at the high-dose level.

Visceral Observations: Not significant. (for pups)

Skeletal Observations:

(From Report)

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Fetotoxicity, as a consequence of severe maternal toxicity, was only observed in the high-dose group as male and female fetal weights were reduced to 71 percent of control values. This fetal growth retardation was associated with a significant increase in variations (primarily localized to the periphery of the fetal skeleton) including: 1) Bipartite, misaligned, and not ossified, centrum/vertebrae, 2) misaligned and not ossified, sternebrae, 3) metacarpals, proximal phalanges, and distal phalanges not ossified in the forepaw, and 4) metatarsals, distal phalanges, not ossified in the hindpaw. There were no compound-related fetal gross, skeletal and visceral malformations. There were no fetal effects at the low or intermediate-dose level.

Discussion:

There are certain discrepancies in this study. The fact that at the high-dose level the toxic symptoms observed in the mothers were severe and significant; including, bleeding (red stains around vulva) and one of the animals, MS 16, had the largest number of pups (19), makes the results questionable.

[illegible]

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
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OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM 23 JUN 1982

Caswell # 125 D

TO: Jackie Dziuban
PM Team No. 25
Registration Division (TS-767)

SUBJECT: Studies submitted by Ciba-Geigy on Terbutryn in response to RPAR.

Four studies were received from Ciba-Geigy (Date of submission; 4/23/82; Accession numbers 247365-247368) in response to FIFRA Section 6 (a) (2). Toxicology Branch has reviewed the studies, and the conclusions are as follows:

Conclusions and Recommendations

(1) Rat-intrasanguine host - mediated assay using S. typhimuruim (Ames) to test for the mutagenicity of GS-14260. This test showed no mutagenic effects but does not meet minimum criteria set by EPA standards.

(2) In vivo hamster cytogenetic study on bone marrow cells showed that GS-14260 caused no chromosome aberrations in this test. Meets minimum criteria set by EPA standards.

(3) Chromosome studies in the germinal epitheluim of male mice suggested that GS-14260 caused no chromosome aberrations in spermatogenia, but the study only of supplementary value and does not meet minimum EPA standards.

(4) Dermal absorption study using ^{14}C-Terbutryn applied to rat skin. Invalid. All supporting experimental results are requested.

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Reviews

1. Host-mediated assay. A rat intrasanguine host-mediated assay using Salmonella typhimurium (Ames) to test for the mutagenicity of GS-14260 (Terbutryn technical) was performed by Ciba-Geigy, Basle, Switzerland, authored by P. Arni and D. Miller, dated 11/20/81. Male albino rats (20-41g; 6 mice per group) that had been fasted for 16 hours were administered Terbutryn in CMC by gavage at levels of 0, 500, 1000 or 2000 mg/kg at 2 hour, 1 hour and immediately before the injection of about 10^{10} bacteria (TA 1535, 1537, 98, or 100) into the tail vein. One hour after injection the rats were sacrificed, and the bacteria recovered from the livers. 10^{7} to 10^{8} bacteria were recovered from each host. 0.2 ml of this suspension was spread on each of 5 plates. The mutation rate (reversion from histidine auxotropy to prototrophy) was not increased in strains TA 1535, 1537 and 98 by administration of Terbutryn, but mutagenic effects were observed in strain TA 100. However, in two replicate experiments no mutagenic effects were seen in TA 100, so the mutagenic effect was not confirmed.

There are a few defects in this study. No postitive controls were mentioned, and this is a serious omission in the host-mediated assay. Positive controls could be performed on 1) the bacteria before, or 2) after injection and recovery from the host, or 3) the host itself by injection of the positive control chemical into the host, followed by injection and recovery of the bacteria. The mutagenic effects of the positive control chemical on the bacteria are then scored by the normal procedure for an Ames' test. Performance of positive control #3 would have been sufficient.

Terbutryn was administered beginning at 2 hours before injection of the bacterial tester strains, and the animals were sacrificed 1 hour after injection of the bacteria. However, the authors did not prove, or even state, that the selected times of administration of test substance and tester strains were the optimal times.

Conclusion:

This study suggests that GS-14260 caused no mutagenic effects, but because of protocol limitations this study does not meet minimum criteria set by EPA standards.

2. In vivo hamster cytogenetics test. Test performed by G. Hool, E. Puri, and D. Muller of Ciba-Geigy, Basle, Switzerland, dated 11/24/81. Chinese hamsters (21-33g) were administered 0, 750, 1500, or 3000 mg/kg of GS 14260 by gavage on each of 2 consecutive days. Four animals per sex per dose were used, and 6 animals per sex were used in the positive and negative control groups. The animals were injected i.p. with 10 mg colcemide/kg 2 hours after the second dose and sacrificed by cervical dislocation 4 hours later. Chromosome preparations were made from bone marrow cells. Two animals per sex per dose were evaluated for chromosome aberrations. One hundred metaphase plates from each animal were analyzed. In two animals in the control groups (0.5% MC + 0.1% Tween 80) an acentric fragment was observed. The test groups showed the following chromosome aberrations: low dose: one break; intermediate dose: one minute chromosome; high dose; none. Cyclophosphamide (64 mg/kg) administered as positive control resulted in 22.0% with chromatid aberrations, 11.8% with chromosome aberrations, and 0.25% of the cells scored revealed pulverizations. Thus GS 14260 was not mutagenic in this test.

Conclusion: GS 14260 caused no chromosome aberrations in Chinese hamsters in this test. Meets minimum criteria set by EPA standards.

3. Chromosome ~~Chronic~~ studies in male germinal epithelium. This study is dated 11/23/81; directed and reviewed by G. Hool and D. Muller, respectively, of Ciba-Geigy, Basle, Switzerland. An NMRI-derived strain of male mice (15 animals per test group, 12 animals in the control group) were administered GS 14260 by gavage (486 and 1458 mg/kg in 20 ml/kg 0.5% CMC--CMC alone was administered as negative control) on 5 consecutive days (days 0-4). The authors state that 4,370 mg/kg was the LD_{50}. On day 5 the mice were given 10 mg/kg Colcemide and sacrificed three hours later. 100 metaphase figures from the testes of each of 8 animals in each group were scored for chromosome aberrations. One animal in the control, and the high dose, group showed one gap; otherwise no chromosome aberrations were observed. Thus no mutagenic effects on spermatogonia were observed.

This study attempts to determine whether or not Terbutryn has heritable mutagenic effects, but is only of supplementary value for the following reasons. Lack of a positive control. Although it may not be necessary to perform a positive control (e.g. mitomycin c, 6-mercaptopurine) during each test series, at least the authors could have provided evidence to show that this procedure as performed in their lab is sensitive to known mutagens.

Lack of experimental detail, particulary in the isolation and preparation of the germ cells for scoring. This test has not been standardized as much as some of the other tests for heritability, and as such needs to be reported more completely. No data presented. Under "Results" only a summary sentence appears in the text, and no Tables or Figures are included. What types of "chromosome and chromatid aberrations" were scored.?

Conclusion:

No chromosome aberrations observed, but without further data the study does not meet minimum criteria set by EPA standards and is only of supplementary value.

4. Dermal absorption of ^{14}C-Terbutryn by rats. Submitted by B. Simoneaux of Ciba-Geigy, Greensboro, N.C.; issue date 4/15/82, Report No. ABR-82016.

This study is inadequately reported, and should be considered invalid until supporting experimental data is received by EPA. There are some important questions that must be answered. For example: (1) How many animals comprised each data point? (2) Please explain the quench correction procedure, i. e., in converting cpm to dpm to ug. (3) Did they (the experimenters) distinguish between "on" as opposed to "in" the skin? The report mentioned in the Methods section that ^{14}C in the acetone washes was counted separetely from that remaining in the washed, excised skin, but the data is not listed under Results and Discussion.

In addition, the following information would be useful to EPA in evaluating this study if the data were obtained. (1) It is unfortunate that the study was termnated at 1 day, particuilarly since ^{14}C-was still being taken up from the skin at 24 hours. In this regard, RBC's appeared to absorb radioactivity throughout the observation period, and one wonders whether or not Terbutryn binds to the RBC. (2) Does Terbutryn bind to fat? Terbutryn is not very soluble in water (about 58 ppm), but very soluble in organic solvents, and one wonders whether it might accumulate in fat. (3) If time points were measured for excretion (urine, feces), please provide the data.

Conclusion:

This study is inadequately reported and is considered invalid until supporting data is received. All supporting experimental results for all animals for all time points is needed, as mentioned above.

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C.T. Chelsvan

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CONFIDENTIAL BUSINESS INFORMATION
DOES NOT CONTAIN
NATIONAL SECURITY INFORMATION (EO 12065)

D 00550
DRAFT

EPA: 68-02-4225
DYNAMAC No. 1-80-A2
June 25, 1986

07-01-

DATA EVALUATION RECORD

TERBUTRYN

Mutagenicity (Chromosomal Damage)--
Micronucleus Test in Chinese Hamsters

STUDY IDENTIFICATION: Hool, G., Langauer, M., and Muller, D. Nucleus anomaly test in somatic interphase nuclei, GS 14 260, Chinese hamsters (test for mutagenic effects on bone marrow cells). (Unpublished study No. 810174 prepared and submitted by CIBA-GEIGY, Ltd., Switzerland; dated November 16, 1981.) Accession No. 261709.

APPROVED BY:

I. Cecil Felkner, Ph.D.
Department Manager
Dynamac Corporation

Signature: _____

Date: _____

1 CHEMICAL: Terbutryn; GS 14 260.

2. TEST MATERIAL: GS 14 260, lot No. 75072, was 96% pure; the physical appearance was not reported.

3. STUDY/ACTION TYPE: Mutagenicity (chromosomal damage)--Micronucleus test in Chinese hamsters.

4. STUDY IDENTIFICATION: Hool, G., Langauer, M., and Muller, D. Nucleus anomaly test in somatic interphase nuclei, GS 14 260, Chinese hamsters (test for mutagenic effects on bone marrow cells). (Unpublished study No. 810174 prepared and submitted by CIBA-GEIGY, Ltd., Switzerland; dated November 16, 1981.) Accession No. 261709.

5. REVIEWED BY:

Nancy E. McCarroll, B.S.
Principal Reviewer
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Date: _____

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~~Marcia Van Gemert~~, Ph.D.
~~EPA Section Head~~
Chief, Mission Support Staff

Signature: _____
Date: _____

7. CONCLUSIONS:

A. Under the conditions of the nucleus anomaly (micronucleus) test, exposure of male and female Chinese hamsters to 750, 1500, and 3000 mg/kg GS 14 260, administered orally on 2 consecutive days, did not significantly increase the incidence of nuclear anomalies (micronuclei). However, the design of the protocol was inappropriate for assessing the full cell cycle for micronuclei induction.

B. The study is unacceptable. I agree! 07-01-85

8. RECOMMENDATIONS:

The following recommendations are made:

1. Retest GS 14 260 in a multiple sampling protocol (48, 72, and 96 hours postexposure) to ensure that maximum sensitivity to detect micronuclei is achieved.

2. Determine the ratio of polychromatic to normochromatic erythrocytes.

3. Report clinical signs.

4. Randomize animals, report identification methods, and code slides.

5. Include a quality assurance statement.

Items 9-10--see footnote 1.

11. MATERIALS AND METHODS (PROTOCOLS):

A. Materials and Methods: (See Appendix A for details.)

1. Test Material: GS 14 260, lot No. 75072, was listed as 96% pure and stable under unspecified conditions for at least 3 years. Dosing solutions of the test material were prepared in 2% carboxymethylcellulose containing 0.1% Tween 80 (CMC+Tw80).

2. Test Animals: Male and female Chinese hamsters (Cricetulus griseus) were used in this study; the source of the animals was not reported. The males weighed between 24-31 g and the females between 20-27 g; the age of the hamsters at the onset of the study was not reported.

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[1]Only items appropriate to this DER have been included.

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3. Animal Maintenance: Acclimation to laboratory conditions and animal housing were not reported. The animals were maintained in an air-conditioned room controlled for temperature (22-24°C), humidity (62-72%), and light (12 hours). Standard diet, NAFAG No. 196, source not specified, and tapwater were provided ad libitum.

4. Assignment to Groups: The method, if any, used to randomize animals was not reported. Animals were identified by individual caging.

5. Rationale for Dose Selection: The report cited the findings of an early study, which indicated that the oral acute LD_{50} in Chinese hamsters of both sexes was >3000 mg/kg. Based on this information 750, 1500, and 3000 mg/kg were selected as the doses for the nucleus anomaly assay.

6. Nucleus Anomaly Test

   a. Test Animals and Compound Administration: Six hamsters (three males and three females) per group were administered the selected concentrations of the test material, vehicle (CMC+Tw80), or the positive control, Endoxan (cyclophosphamide, 128 mg/kg), in two single applications separated by 24-hour intervals. Dosing solutions were prepared to yield volumes of 20 mL/kg.

   b. Animal Sacrifice/Bone Marrow Harvest: Twenty-four hours after the second application of the ~~test material~~, vehicle, or positive control, the animals were sacrificed by cervical dislocation. Bone marrow cells were harvested from both femurs by aspiration into 0.5 µL rat serum. Aspirates were mixed, dropped onto slides, and air dried. Prepared slides were stained in undiluted May-Grünwald solution and diluted May-Grünwald (1:1 in H_2O), counterstained in Giemsa, ~~and mounted~~. The report did not specify whether the sides were coded.

   c. Slide Analyses: One thousand bone marrow cells per animal were scored for the following nuclear anomalies: single Jolly bodies, fragments of nuclei in erythrocytes, micronuclei in erythroblasts, micronuclei in leucopoietic cells, and polyploid cells.

7. Statistical Analysis: The data were analyzed for significance using the ~~Chi-square~~ test. Although the p value was not specified, it was assumed that the finding would be significant at p ~~<0.05 since the~~ positive control was reported to be significant at this level.

8. Evaluation Criteria: Criteria for a positive response, the validity of the assay, or the biological significance of the findings were not present.

B. Protocol: A protocol was not presented.

12. REPORTED RESULTS:

~~Nucleus Anomaly~~ Assay: A supplement to the report stated that clinical signs were not recorded in the nucleus anomaly assay. Similarly, the ratio of polychromatic erythrocytes to normochromatic erythrocytes was not calculated. No significant increase in the ~~percentage of~~ cells with nuclear anomalies in either male or female Chinese hamsters occurred following two gavage administrations of 750, 1500, or 3000 mg/kg GS 14 260. Cyclophosphamide, 128 mg/kg (po x 2), induced significant ($p < 0.05$) increases in the total percent of cells wth nuclear anomalies for both sexes. Representative results are shown in Table 1.

13. STUDY AUTHORS' CONCLUSIONS/QUALITY ASSURANCE MEASURES:

A. The authors stated, "It is concluded that under the conditions of this experiment, no evidence of mutagenic effects was obtained in Chinese hamsters treated with GS 14 260."

B. A quality assurance statement was not present; however, a signed statement dated November 7, 1985, ~~indicated~~ that the study was conducted in compliance with the FIFRA Good Laboratory Practice Standards.

14. REVIEWERS' DISCUSSION AND INTERPRETATION OF STUDY RESULTS:

We assess that the methods used to determine micronuclei induction (cell harvest ~~approximately 18~~ hours after the first test material administration) were inadequate to detect compound-related effects on the majority of ~~cells in first~~ and second division. Although micronuclei do not immediately vanish, cells with micronuclei will be progressively diluted in any cell population that continues to proliferate. Also, cells affected by the second dosing would continue to produce micronuclei, whose appearance might be delayed and detected only after 72 or even 96 hr. Thus, the ability of this method to detect subtle increases in micronuclei at different stages in the cell cycle is doubtful. Salamone et al.[2] have shown that

[2] Salamone, M. J., Heddle, J. A., Stuart, E., and Kate, M. Towards an improved micronucleus test: Studies on three model agents, Mitomycin C, cyclophosphamide and dimethylbenzanthracene, Mutat. Res. 74(1980): 347-356.

no single sampling time gives maximum sensitivity for all chemicals, even after two compound treatments, and recommend using multiple sampling intervals (48, 72, and 96 hours) to increase assay sensitivity.

Item 15--see footnote 1.

16. CBI APPENDIX: Appendix A, Material and Methods, CBI pp. 20-21.

TABLE 1. Representative Results of the Nucleus Anomaly Test (Micronucleus Assay) in Chinese Hamsters with GS 14 260

Substance	Dose[a] (mg/kg)	No. of Animals Analyzed per Group	No. of Cells Analyzed per Group	Average Percent Cells with Nuclear Anomalies[b]					
				Jolly Bodies	Nuclear fragments in Erythrocytes	Micronuclei in Erythroblasts	Micronuclei in Leucopoietic Cells	Polyploid Cells	Total
Vehicle Control									
2% Carboxymethyl-cellulose + 0.1% Tween 80		3 M	3000	0.03	0	0	0	0	0.03
		3 F	3000	0.06	0	0	0.03	0	0.09
Positive Control									
Endoxan (cyclophosphamide)	128	3 M	3000	9.9	1.3	1.7	0.6	0.03	13.4*
		3 F	3000	7.8	1.2	1.4	0.2	0	10.7*
Test Material									
GS 14 260	3000[c]	3 M	3000	0.06	0	0	0	0	0.06
		3 F	3000	0.13	0	0	0	0	0.13

[a] Two gavage administrations separated by 24 hours; sampling was at 24 hours after the second dosing only.

[b] Averaged by our reviewers.

[c] Similar results were obtained at the low- (750 mg/kg) and mid- (1500 mg/kg) doses; therefore, the data from the highest (3000 mg/kg) dose were selected as representative.

*Significantly different from control value ($p < 0.05$) by the Chi square test.

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PROCEDURE

1. Data on the animals used.

Chinese hamsters (Cricetulus griseus) of either sex (♀:♂ = 1:1)(weight ♀♀ 20-27 g, ♂♂ 24-31 g) were used. Standard diet: NAFAG No.924. Tap water ad libitum. The animals were kept in an air-conditioned room at a temperature of 22-24°C and a relative humidity of 62-72%. The room was illuminated for 12 hours daily. Identification of the animals by individual caging.

2. Treatment schedule.

The preparation was administered orally to groups of 6 female and 6 male animals each. Treatment consisted of daily one application on 2 consecutive days. 24 h after the second application the animals were sacrificed by dislocation of the cervical vertebrae.

a) GS 14 260: 750, 1500 and 3000 mg/kg in 20 ml/kg 0.5% aqueous solution of Sodium-Carboxymethylcellulose (CMC) plus 0.1% Tween 80 (Tw 80). *)

b) Cyclophosphamide (ENDOXAN®): 128 mg/kg in 20 ml/kg CMC + Tw 80 (positive control).
Manufacturer of ENDOXAN®: ASTA-Werke, Germany.

c) 20 ml CMC + Tw 80/kg (negative control).

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3. Preparation of bone marrow.

Bone marrow was harvested from the shafts of both femurs. In a siliconized pipette filled with approx. 0.5 µl rat serum the bone marrow was drawn up. In order to receive a homogeneous suspension the content of pipette was aspirated gently about three times. Small drops of the mixture were transferred on the end of a slide, spread out by pulling it behind a polished

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*) The oral acute LD_{50} was found to be >3000 mg/kg in Chinese hamsters of either sex (cf. Lab.Report: GU 2.1, dated March 23, 1981).

cover glass and the preparations were air-dried. Three hours later, the slides were stained in undiluted May-Grünwald solution for 2 min then in May-Grünwald solution/water 1/1 for 2 min and then in Giemsa's, 40% for 20 min. After being rinsed in methanol 55% for 5-8 sec and washed off twice in water, they were left immersed in water for approx. 2 min. After rinsing with distilled water and air-drying, the slides were cleared in Xylol and mounted in Eukitt.

4. Scoring of the slides.

The slides of three female and three male animals each of the negative control group, the positive control group and of the groups treated with various doses of GS 14 260 were examined. 1000 bone marrow cells each were scored per animal and the following anomalies were registered:

a) Single Jolly bodies, b) fragments of nuclei in erythrocytes, c) micronuclei in erythroblasts, d) micronuclei in leucopoietic cells, e) polyploid cells.

5. Statistics.

The significance of difference was assessed by χ^2-test.

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Data Evaluation Report

Compound Terbutryn

Citation

Dermal Absorption of Terbutryn in Rats, T. Murphy & B. Simoneaux, Biochemistry Dept, Agricultural Div., Ciba-Geigy, Report # ABR-86017, 2/28/86. Acession No.261708.

Reviewed by Robert P. Zendzian PhD 4/29/86
Pharmacologist

Core Classification Aceptable

Conclusion

Significant quantities of terbutryn were absorbed dermally at all doses and time intervals. Representative percent absorptions for 10 hours exposure were 22.02, 4.44 and 2.25 for doses of 0.05, 0.5 and 5.0 mg/10cm^2. Animals washed at ten hours and maintained for an additional 48 hours showed total absorptions of 34.48, 12.26 and 8.25 percent of the respective doses.

Materials

Terbutryn, uniformly labeled with ^{14}C in the triazine ring. Specific activity 13.1 uCi/mg for the low and mid doses, 1.31 uCi/mg for the high dose. Radioactive purity 99%

Male Harlan Sprague-Dawley rats 200-250 gms from Madison, Wisconsin.

Methods

Dose preparation. "The low dose was prepared by dry mixing throughly 5.0 mg of ^{14}C-terbutryn with 2.5 mg of blank formulation (WP) and then suspending the mixture with 5.0 ml of deionized water. The middose was prepared by dry mixing throughly 25 mg of ^{14}C-terbutryn and 12.5 mg of blank formulation (WP). The mixture was suspended in 2.5 ml of deionized water. The resultant mixtures simulate a typical 80W formulation."

"The high dose was prepared by dry mixing throughly 62.5 mg of blank formulaion (WP), 225 mg of cold terbutryan and 25 mg of ^{14}C-terbutryn and then suspending the mixture in 2.5 ml of deionized water."

Animal preporation. The back of the male rats was shaved and washed with acetone 24 hours prior to treatment. A 4.0 by 2.5 cm area, 10 cm^2, was marked as the application site. The rear legs of the rats were restrained to prevent scratching of the application site.

Site protection. In the 'Preliminary Study' the application site was left completely uncovered. In the main study the site was covered by the device described below.

Dose application. "Fifty microliters of an aqueous suspension containing either the low (0.05 mg/rat), mid (0.5 mg/rat) or high (5.0 mg/rat) dosage levels was applied with a 50 ul Hamilton Syringe equipted with a Teflon tip coated needle and plunger assembly. The tip of the needle was used to uniformly spread the suspension over the entire treatment area. The amount of ^{14}C-terbutryn applied to the rat was calculated by radioassay of 50 ul of ^{14}C-te butryn delivered from the same syringe."

Site protection main study. "After dosing, the treated area was allowed to air dry for five to ten minutes and the entire treated area was enclosed by a nonocclusive covering consisting of Stomanesive® (Squibb Corporation), filter paper and an aluminum bridge (Figure 2). Skin-bond cement (Pfitzer Corp.) was evenly spread around a one centimeter border surrounding the dose area. The Stomanesive was placed on top of the glued area to form a "well" surrounding the treated skin area. The aluminum foil bridge, which was slightly curved to elevate the filter paper, was glued to each side of the Stomanesive in order to cross directly over the center of the dose area. The entire area was "covered" by Whatman No. 1 filter paper which was glued to the Stomanesive barrier."

Experimantal Design

Preliminary study. Four rats were dosed with 0.5 mg/rat and exposed for 10 hours.

Main study. Rats were dosed and exposed as follows,

Dose mg/rat	Exposure Period (hours) 2	4	10	24	50*
0.05	4	4	4	4	4
0.5	4	4	4	4	4
5.0	4	4	4	4	4

*10 hours after treatment the application site was washed as described below and the rats placed in clean metabolism cages for an additional 48 hours.

Treated rats were placed in individual metabolism cages and feces and urine collected for the exposure period. At the end of the exposure period the cages were washed with acetone and water and the rats were sacrificed. The protective appliance was removed and its components seporated for analysis. The application site was washed with Dove liquid in water (20ml : 1 liter) and then with deionized water using gauze squares. Two skin samples were removed, I the dosed area and II the surrounding area covered by the appliance. A blood sample was collected and the carcass retained for analysis.

The following samples were analyzed for each treated rat. Skin I, skin II, blood, carcass, cage wash, soap rinse, water rinse, Stomahesive rinse, paper rinse, bridge rinse, paper and gauze squares.

Results

Preliminary study

In the preliminary study the application site was unprotected and the results, from Table 1, show significant "fall off" of the material during the 10 hour exposure period.

Table A. Mean percent recovery, dose 0.5 mg/rat, exposure 10 hours. Four rats.

Samples	Mean
Plasma	00.20
RBC	00.40
Carcass	46.06
Skin	31.85
Skin Wash	12.78
Cage Wash	10.40
Urine	6.34
Feces	00.08

Percent absorbed 53.08 (plasma, RBC, carcass, urine and feces)
Percent skin 31.85
Percent unabsorbed 23.18 (skin wash and cage wash)

Main study

Table B. Distribution of the applied dose as mean percent of dose.

Dose mg/rat	Fraction	Time of exposure (hours) 2	4	10	24	58*
0.05	Absorbed	10.52	9.58	22.02	31.48	34.48
	Skin	12.49	21.03	21.25	22.40	4.47
	Unabsorbed	68.00	66.22	70.47	48.82	55.25
	Total	91.01	96.83	113.78	102.70	94.29
0.5	Absorbed	1.42	3.06	4.44	14.25	12.26
	Skin	20.03	7.34	22.25	12.66	16.05
	Unabsorbed	93.93	82.54	75.45	84.38	93.53
	Total	115.38	92.94	102.14	111.29	121.84
5.0	Absorbed	1.34	2.48	2.25	4.80	8.25
	Skin	9.83	14.49	7.16	14.66	3.37
	Unabsorbed	61.66	54.12	80.84	51.84	74.73
	Total	72.39	81.09	90.25	71.30	86.33

*Application site washed at 10 hours and exposed for an additional 48 hours.

Discussion

This study shows clearly the necessity of protecting the application site in order to retain material. Comparing the unprotected animals with the appropriate protected animals, it appears that 12 times as much material was absorbed by the unprotected rats. This obviously represents material which was mainly eaten by the rats by licking the application site and/or by eating material deposited on the sites of the metabolism cage from the rats' back.

The study shows that terbutryn is readily and rapidly absorbed. Of particular importance is the quantity remaining on the skin after washing with soap and water. Comparision of the date from the group that was sacrificed at 10 hours and the date from the group that was washed at 10 hours and then followed for an additional 48 hours showes that a major portion of this residual material must be considered as available for absorption.

3. Peer Review Committee Members in Absentia: (Committee members who were not able to attend the discussion; signatures indicate concurrence with the overall conclusions of the Committee.)

Anne Barton

Diane Beal

Richard Hill

Stephen Johnson

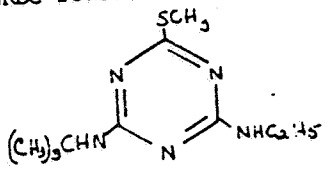
B. Material Reviewed:

The material available for review consisted of DER's on rat (Charles River CD) and mice (CD-1) oncogenicity studies of Terbutryn.

C. Background

Terbutryn is a selective herbicide registered for postemergence on winter wheat and barley; preemergence and preplant incorporated on grain sorghum; and preemergence and postemergence on fallow. It is structurally similar to several other triazine herbicides: simazine, cyanazine, propazine and atrazine.

A registration standard is presently being generated on terbutryn and it is also the subject of an NRDC reassessment and Data-Call-In.



Terbutryn
(2-t-butylamino-4-ethylamino-6-methylthio-s-triazine)

D. Evaluation of the Evidence:

1. Mouse Oncogenicity Study of Terbutryn:

a. Data Considered:

Two-hundred-forty male and 240 female Charles River CD-1 mice were initiated in this study. Sixty male and 60 female mice were placed in one of four groups: 0, 3, 1000, or 3000 ppm terbutryn in the diet. The study was conducted by International Research and Development Corporation (IRDC). The test material was terbutryn technical (ARS No. 2046/76; Batch No. FL-761552, white powder).

No treatment-related effects were seen on general behavior, appearance, body weight gain, food consumption or survival. No-treatment related non-neoplastic effects were noted in any of the male mice. Slight increases in amyloidosis were seen in various organs/tissues of female mice as well as an increase in brown pigment in the cervical lymph node. The incidence of these lesions is summarized in the following table.

Incidence of Non-Neoplastic Lesions in Female Mice

	Group (ppm)			
Non-neoplastic lesion	0	3	300	3000
Thyroid amyloidosis	5/58	0/0	0/0	13/57
Parathyroid amyloidosis	3/32	0/0	0/0	8/30
Adrenal amyloidosis	11/60	0/0	0/0	22/56
Stomach amyloidosis	1/60	0/3	0/9	6/60
Kidney amyloidosis	13/60	11/19	12/19*	28/58
Lymph Node, Cervical brown pigment, reticulo-endothelial cells	8/57	0/1	0/2	26/55

* From the report it was difficult to decipher this number.

No neoplastic lesions were seen that could be considered to be treatment-related.

b. MTD Consideration:

The Committee concurred that there was no evidence of oncogenicity due to terbutryn seen in the CD-1 mouse; however, they felt that the MTD was not reached in this study. In a four week mouse study the NOEL was determined to be 3000 ppm based upon decreased weight gain, corneal opacity and eccentric pupils seen at 10,000 ppm. A more appropriate high dose for this study was considered to be 7500 ppm. The Committee decided that a repeat study was not required since 1) the highest dose tested was close to one half the expected MTD, 2) structurally related triazines have not been found to be oncogenic toward CD-1 mice (see the section on Structure Activity Relationship under E. Additional Non-oncogenic Data. of this report), and 3) there is no evidence that terbutryn is mutagenic.

2. Rat Oncogenicity Study of Terbutryn:

a. Data Considered:

Two hundred-sixty male and 260 female weanling Charles River CD rats were initiated in this study which was conducted by IRDC (Report dated March 27, 1980). They were placed in one of the groups as depicted in the table below.

Group (ppm)	Number of Rats/Group Male	Female
0	70	70
2	60	60
300	60	60
3000	70	70

Ten additional rats were placed in the control and high dose groups. Five of these rats of each sex were killed and necropsied at 12 months. The other five per sex of the high dose group were placed on control diet at the twelve month period and were killed and necropsied, along with the remaining 5 control rats, four weeks later. The test material was terbutryn technical (ARS No. 2046/76, Batch No. FL761552).

Survival was unaffected by treatment. Body weight gain for both males and females was significantly reduced in the high dose group (20% for males and 30% for females). Erythrocyte and hemoglobin values were significantly decreased at 18 months for all dosage groups and SGOT and alkaline phosphatase values were significantly elevated at the 3 and 6 month and at 12, 18, and 24 months in the high dose group, respectively.

Several variations in absolute and relative organ weights were seen at the highest dose tested. These included a statistically significant decrease in absolute spleen weights in male and female rats, increase in relative liver, kidney, brain and thyroid weights in males and females, increase in absolute and relative testes weights in males, increase in relative ovarian and adrenal weights in females and an increase in relative but decrease in absolute heart weights in male and female rats.

The incidence of neoplastic lesions seen in this study can be found in the following two tables.

Incidence of Mammary Tumors in Female Rats

	Control	Low	Mid	High	Sign. (p)[1]
# animals with adenomas[2]	6/57(10.5)[3]	9/58(15.5)	8/58(13.8)	13/55(23.6)	0.055
# animals with adeno-carcinomas[2]	15/57(26.3)	8/58(13.8)	7/58(12.1)	20/55(36.4)	0.170
# animals with fibro-adenomas[2]	12/57(21.1)	15/58(25.9)	20/58(34.5)	18/55(32.7)	0.120
# animals with adenomas and/or adenocarinomas[4]	18/57(31.6)	17/58(29.3)	13/58(22.4)	28/55(50.9)	0.03
# animals with adenomas and/or adenocarcinomas and/or fibroadenomas[4]	24/57(42.1)	29/58(50.0)	29/58(50.0)	34/55(61.8)	0.03

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1 Derived from Fishers Exact test comparison of control and high dose groups.
2 These animals could have other types of mammary tumors as well.
3 Numbers in parentheses indicate percentage incidence.
4 There is no duplication of animals in these numbers. These are numbers for individual animals.

Incidence of Other tumors and Pertinent Non-Neoplastic Lesions in Male and Female Rats.

Organ/Tissue	Control	Low	Mid	High	Sign. (p)*
			Male		
Thyroid					
follicular cell					
adenoma	1/59(1.7)**	0/59(0)	0/60(0)	6/57(10.5)***	
carcinoma	0/59(0)	0/59(0)	1/60(1.7)	3/57(5.3)	
aden. + carcin.	1/59(1.7)	0/59(0)	1/60(1.7)	9/57(15.8)	0.007
hyperplasia	0/59(0)	0/59(0)	0/60(0)	2/57(3.5)	
Testes					
interstitial cell					
adenoma	13/59(22.0)	11/60(18.3)	14/60(23.3)	23/57(40.4)	0.026
hyperplasia	0/59(0)	2/60(3.3)	3/60(5.0)	0/57(0)	
			Female		
Liver					
hepatocellular					
adenoma	3/57(5.3)	2/60(3.3)	3/59(5.0)	12/56(21.4)	
carcinoma	2/57(3.5)	0/60(0)	0/59(0)	4/56(7.1)	
aden. + carcin.	5/57(8.8)	2/60(3.3)	3/59(5.0)	16/56(28.6)	0.006
focal cytomegaly	9/57(15.8)	13/60(21.7)	9/50(18.0)	18/56(32.1)	

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* Derived from Fishers Exact test comparison of control and high dose groups.
** Number in parentheses indicates the percentage incidence..
*** A thyroid adenoma was also seen in a male rat sacrificed at 12 months. This tumor is not included in this figure.

b. Historical Control Information:

Historical control data from IRDC on Charles River CD rats were available from the FDA files. These studies were initiated in the late 1970's and were designed with two control groups per study. The total number of studies the data were derived from was six. These data are summarized along with data from the control and high dose groups from the terbutryn study on the following page. Only these two groups were considered since only a high dose effect was apparent.

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IRDC Historical Control Data Compared to Tumor
Data Obtained in the Two Year Terbutryn Rat Study

Tumor Type	Historical Control Data Incidence	%	Range(%)	Terbutryn Data Control	High Dose
			Males		
Testes					
interst. cell and/or Leydig cell tumors	49/739	6.6	0-14	13/59(22.0)	23/57(40.4)
Thyroid					
follicular cell					
adenoma	14/731	1.9	0-3.6	1/59(1.7)	6/57(10.5)
carcinoma	2/731	0.27	0-1.8	0/59(0)	3/57(5.3)
combined[1]	16/731	2.19	-	1/59(1.7)	9/57(15.8)
			Females		
Liver[2]					
neoplastic nodules	15/750	2.0	0-8.6	3/57(5.3)	12/56(21.4)
hepatocellular carcinoma	3/750	0.4	0-1.7	2/57(3.5)	4/56(7.1)
combined*	18/750	2.4	-	5/57(8.8)	16/56(28.6)
Mammary Gland[3]					
benign tumors	415/733	56.6	31.8-82.6	14/57(24.6)	26/55(47.2)
malignant tumors	120/733	16.4	1.7-23.7	15/57(26.3)	20/55(36.3)
combined[1]	-	-	-	24/57(42.1)	34/55(61.8)

[1] The historical control data for liver and thyroid adenomas and carcinomas was combined with the assumption that no one animal had both an adenoma and a carcinoma. This assumption could be incorrect so that the combined numbers could be slightly higher than in actuality. The historical control data for mammary tumors was not combined for consideration since the likelihood of one animal having multiple types of mammary tumors is high. Individual animal data was used from the terbutryn study and, therefore, no animal was counted twice in the combined incidence data presented.

[2] In the terbutryn study the data represent the number of tumors diagnosed as adenomas. No neoplastic nodules were diagnosed in the study.

[3] In the terbutryn study the only benign mammary tumors were adenomas and fibroadenomas; the only malignant mammary tumors were adenocarcinomas.

In all cases, except for benign mammary tumors, the tumor incidence in the high dose group of the terbutryn study exceeded the upper range of the historical control data.

c. MTD Considerations:

The MTD was reached in this study as evidenced by a 20% depression in body weight gain in males and 30% in females at 3000 ppm. In addition, SGOT and alkaline phosphatase values were significantly elevated and hemoglobin and erythrocyte values were significantly decreased at several time points during the study in the high dose group.

E. Additional Non-Oncogenic Information:

1. Metabolism:

Metabolism studies have been conducted in the rat using both ring- and methylthio-^{14}C-labelled terbutryn. Eighty-five percent of the ring-labelled dose was excreted within 72 hours in urine and feces. Sixty-two percent of ^{14}C-methylthio-labelled terbutryn was recovered within 72 hours in expired CO_2.

	Percent of ^{14}C-label	
	Rat[a]	Rat[b]
CO_2	62.4	-
urine	11.1	39.7
feces	4.3	46.1
carcass	17.3	2.6
total	95.1	88.4

[a]methylthio-^{14}C-labelled terburyn
[b]ring-labelled ^{14}C-labelled terbutryn

Identified metabolites of terbutryn found in rat feces and urine are shown in Figure 1. The major pathways for metabolism are desulfuration, N-deethylation and S-demethylation.

2. Non-Oncogenic Toxicological Effects:

The acute oral LD_{50} for rats is 2.5 g/kg. The acute dermal LD_{50} for rabbits is >2000 mg/kg. Terbutryn induces corneal opacity.

Subchronic toxicity studies on terbutryn have not been done in the rat or dog.

In a 6 month beagle dog study the NOEL was determined to be 10 mg/kg/day based upon mucosal thickening of various segments of the small intestine and submucosal lymphoid hyperplasia in the pyloric region of the stomach at 25 and 50 mg/kg/day. One dog placed in a four week recovery group also had submucosal lymphoid hyperplasia.

In a three generation reproduction study in Charles River-CD rats the NOEL was 300 ppm. At 3000 ppm, decreased mean body weights and food consumption values were found for the F_0, F_1 and F_2 parents as well as decreased pup

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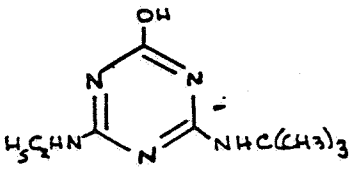
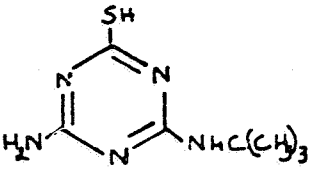
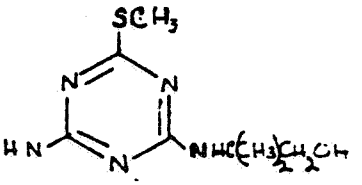
Figure 1. Metabolites of Terbutryn Identified in Rat Urine and/or Feces



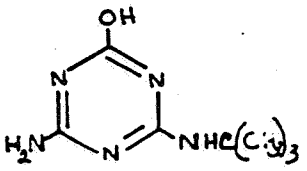
Urine 12% *
Feces 1% *



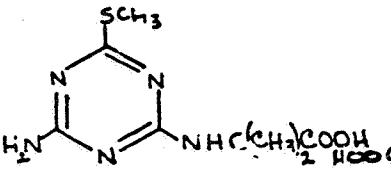
Urine 11% *



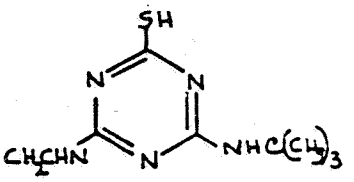
Urine 2% **



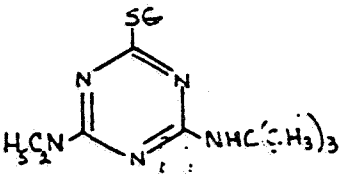
Urine 14%*
Feces 5%*



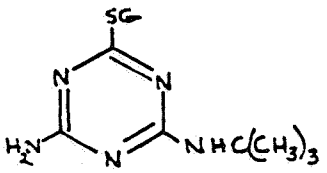
Urine 4%**



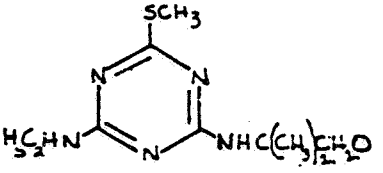
Urine 7%*



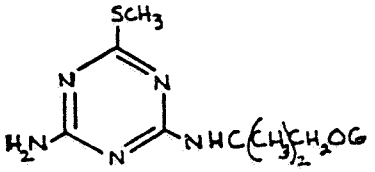
Urine 9%*



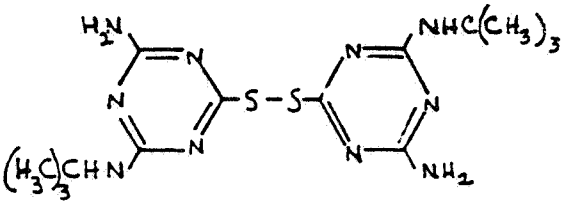
Urine 2% *



Urine 1% **



Urine 4% *
Urine 3% **



Urine 1% **

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* Isolated from rat given ring-labelled terbutryn
**Isolated from rat given Methylthio- labelled terbutryn

G = glucuronide

weights in all generations at lactation day 21.

Teratology studies on terbutryn have been conducted in rats and rabbits. Terbutryn was not teratogenic to either species. The NOEL for maternal toxicity in the rabbit was 10 mg/kg and 50 mg/kg for the rat, based upon decreased food consumption, increased food index and decreased body weight gain and stool changes in rabbits at 50 mg/kg and increased mortality, salivation, urine staining, blood discharge and weight loss at 500 mg/kg in the rat. The NOEL for fetotoxicity in the rat and rabbit was 50 mg/kg based upon reduced ossification and misalignment of the sternebrae and centrum vertebrae, reduced ossification of the metacarpals, proximal phalanges and distal phalanges of the forepaw and reduced ossification of the metacarpals and distal phalanges of the hindpaw in rats at 500 mg/kg and reduced ossification of sternebrae in rabbits at 75 mg/kg.

3. Mutagenicity

The available mutagenicity data was presented as summarized in the following table.

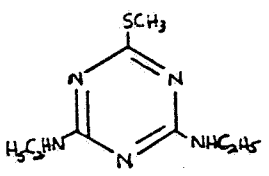
Test	Core Classification	Result	Comments
in-vivo cytogenetics in hamsters	acceptable	negative	no chromosomal aberrations in bone marrow up to 3000 mg/kg
Ames Salmonella	acceptable*	negative	negative up to solubility limit
Micronucleus	acceptable*	negative	negative at 3000 mg
Sister Chromatid Exchange	uninterpretable*	-	several reporting deficiencies

* These studies are still under review by Dynamac. The results given are from a preliminary screen.

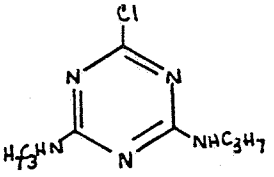
Terbutryn was not found to be mutagenic in any of the assays reported.

4. Structure Activity Relationship:

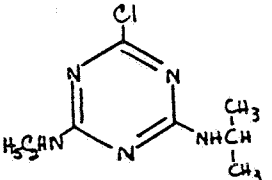
Terbutryn is structurally related to several other triazine herbicides, namely simazine, cyanazine, atrazine, and propazine.



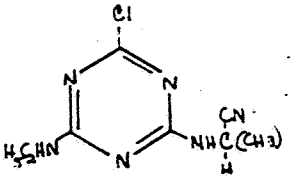
Simazine



Propazine



Atrazine



Cyanazine

Oncogenicity studies have been conducted on all of these triazines but many of them are deficient and cannot be used for a weight-of-the-evidence evaluation.

a. Simazine:

Simazine is rapidly metabolized in the rat. Eighty-six percent of the labelled compound is excreted within 48 hours in the urine and feces. Characterization of metabolites has not been done. Oncogenicity studies are presently underway.

b. Cyanazine:

In rats, 87.84% of labelled cyanazine is eliminated within 4 days, 41.63% in urine and 47.21% in feces. Five and one third percent remains in the carcass. The major metabolic pathways in the rat and cow are dechlorination and deethylation.

Cyanazine did not produce chromosomal aberrations in bone marrow of mice. No other studies were available to evaluate the mutagenic potential of this compound.

Cynazine did not appea. to be oncogenic to CD mice. Studies adequate to determine the oncogenic potential of cyanazine in rats were not available. A new study in the rat has just been initiated.

c. Atrazine:

Atrazine is rapidly eliminated in the rat. Sixty-seven to 72% of the label is excreted within 48 hours in urine and feces. Identification of metabolites has not been done.

Atrazine is not mutagenic in the Ames Salmonella assay or the rec assay using H 17 Rec^+ and M 45 Rec^- strains of Bacillus subtilis. These were the only assays available to judge the mutagenic potential of atrazine.

A study adequate to evaluate the oncogenicity of atrazine in mice has not been done. A 13 month interim report is available on a chronic toxicity/oncogenicity study of atrazine in Charles River Sprague-Dawley rats. An increased incidence of mammary gland adenocarcinomas was reported as follows: 0/22 (control), 1/5 (10 ppm), 1/1 (70 ppm), 0/5 (500 ppm) and 8/25 (1000 ppm). Preliminary data have also been reported for terminal sacrifice. The incidence of adenocarcinomas of the mammary gland was: 15/66 (control), 15/64 (10 ppm), 26/68 (70 ppm) 27/65 (500 ppm) and 35/64 (1000 ppm).

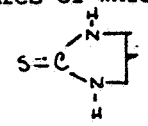
d. Propazine:

Forty-two percent of a ^{14}C-propazine dose was eliminated in the urine of rats and 28% in the feces.

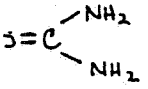
No mutagenicity information was available to the Committee for evaluation.

Propazine was negative for oncogenicity in the mouse (CD-1) but caused a statistically significant increase in mammary tumors in female CD rats. The number of mammary tumor bearing rats was 27/56 (0 ppm), 33/57 (30 ppm), 32/60 (100 ppm) and 39/55 (1000 ppm). The increase in tumors in the high dose group was significant at $p<0.05$.

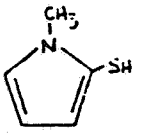
Terbutryn is also structurally similar to several thyroid inhibitors, namely thiourea, ethylene thiourea, thiouracil, propylthiouracil and methimazole, structures of which are shown below.



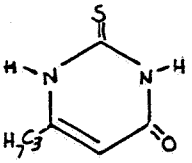
Ethylene Thiourea



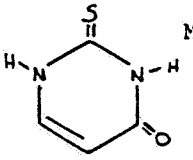
Thiourea



Methimazole



Propylthiouracil



Thiouracil

Chronic studies on thiourea have shown that it induces hepatomas and thyroid enlargement in rats. Thyroid neoplasia was not observed. In another study in rats thiourea was reported to induce malignant tumors of the face. Thiourea was negative for mutagenicity in the Ames Salmonella assay for tester strains TA 1530 and 1538 but positive in TA 100, negative for sex-linked recessive lethals in Drosophila and for UDS in rat hepatocytes, and positive for mutagenicity in S. cerevisiae D_6.

Ethylene thiourea induces hepatomas in two strains of mice (C57Bl/6XC3H/Anf and C57Bl/6X AKR) and thyroid tumors in Charles River CD rats. It has been reported to be weakly positive in the Ames Salmonella assay but these results were not reproducible. Ethylene thiourea has also been reported to be weakly mutagenic toward S. cerevisiae, and to cause an increase in chromosomal aberrations in the bone marrow of mice. It was negative for sex-linked recessive lethals in Drosophila and in the dominant lethal test in mice.

Methimazole, propylthiouracil and thiouracil all induce thyroid tumors in rats. Propylthiouracil also induces pituitary adenomas in mice and thiouracil induces hepatomas in C3H mice.

F. Weight of Evidence Consideration:

The Committee considered the following facts regarding the toxicology data on Terbutryn to be important in a weight-of-the-evidence determination of oncogenic potential.

1. When administered in the diet to female Charles River CD rats terbutryn induced a statistically significant increase in combined mammary gland adenomas and adenocarcinomas and in combined hepatocellular adenomas and carcinomas. In males, terbutryn induced an increase in combined thyroid follicular adenomas and carcinomas and in testicular interstitial cell adenomas.

2. The highest dose level of terbutryn administered to Charles River CD rats reached the maximum tolerated dose in both male and females. This was evidenced by a 20% body weight depression male rats and a 30% body weight depression female rats.

3. Historical control information from the performing laboratory (IRDC) provided additional evidence as to the biological significance of the

increased incidence of thyroid, testicular and liver tumors in the terbutryn treated rats. In all cases, except for benign mammary tumors, the tumor incidence in the high dose group of the terbutryn study exceeded the range of the historical control data.

4. Structure activity information on two structurally related triazine herbicides, propazine and atrazine, provided support for the association of mammary tumors with this class of chemicals. Propazine when administered in the diet to female CD rats, induced a statistically significant increase in mammary gland tumors and atrazine induced a statistically significant increase in mammary gland adenocarcinomas in female Sprague-Dawley rats. Furthermore, four stucturally similar thyroid inhibitors, ethylene thiourea, methimazole, propylthiouracil and thioruacil are known to induce thyroid neoplasia in rats.

5. Available information on the mutagenicity of terbutryn indicates that it is not genotoxic; however, only four assays were available for evaluation.

6. Terbutryn was not oncogenic to the CD-1 mouse.

G. Classification of Oncogenic Potential:

The Committee concurred that the classification of terbutryn, considering all of the available information, should be category C since the chemical produced 1) a marginal response in a tissue (mammary gland) known to have a high and variable background rate, 2) an increase in combined benign and malignant tumors (testicular, thyroid and liver) with an agent showing no response in a variety of short-term tests for mutagenicity (limited, but negative mutagenicity data were available), and 3) a tumor response only in one species. The Committee also considered a category B-2 classification for terbutryn since tumors were produced at multiple sites and since positive, but not conclusive, structure activity relationship (SAR) data were available on other triazines. The SAR data was not considered conclusive since, for propazine, historical control data on the mammary tumors seen in the study is still outstanding and, for atrazine, only a preliminary report on the incidence of mammary tumors was available and the final report has not been evaluated. In addition four thyroid inhibitors which are stucturally similar to terbutryn, are known to induce thyroid neoplasia. The Committee felt that a category C classification was most appropriate but that positive information in the area of mutagenicity for terbutryn and/or mutagenicity and oncogenicity for other structurally related triazines could raise terbutryn to category B-2 classification. In light of this possibility, the Committee decided that a quantitative estimation of the oncogenic potential for humans should be developed.

The dosage selection for both studies was criticized by the Committee. It appeared that the studies were designed such that a dose-response would not be seen except at the highest dosage level which is exactly what occured in the two year rat study.

There was also a discussion on the biological significance of the mammary tumors. The Committee felt that if this were the only tumor type seen, the weight-of-the-evidence would not support the positive oncogenicity of terbutryn because of the high and variable rate of these tumors. However, considering potential structure activity support for mammary tumors from propazine and atrazine oncogenicity studies in the same strain of rat, the Committee decided that in the

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case of terbutryn these tumors were not only statistically but biologically significant, i.e. related to compound administration.

The appropriate combination of mammary tumors for statistical analysis was also considered. NTP in "Report of the NTP Ad Hoc Panel on Chemical Carcinogenesis Testing and Evaluation" does not recommend combining fibroadenomas and adenocarcinomas of the mammary gland. The Committee decided that the evidence for such a decision is equivocal and they elected to combine all mammary tumors for statistical analysis, in addition to combining adenomas and adenocarcinomas.

Questions arose to the possibility of a hormonal mechanism for the induction of the tumors seen in this study, especially in light of the Agency's developing policy on a threshold for thyroid neoplasia. The Committee concluded that sufficient information is not available on the mechanism of oncogenicity of terbutryn to consider a threshold for its neoplastic effects.