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

Office of Prevention, Pesticides
and
Toxic Substances

17PP

MEMORANDUM

SUBJECT: **TERBUFOS**: Toxicology Chapter for RED

FROM: Linda L. Taylor, Ph.D. 8/26/99
Reregistration Branch I,
Health Effects Division (7509C)

TO: William Hazel
Reregistration Branch I,
Health Effects Division (7509C)

Registrant:	American Cyanamid Company
Chemical:	S-[[(1,1-dimethylethyl)thio]methyl]O,O-diethyl phosphorodithioate [IUPAC}; S-(*t*T-butylthio)methyl O,O-diethyl-phosphorodithioate; S-tert-butylmercaptomethyl O,O-diethyl dithiophosphate,
CAS No.:	13071-79-9
Caswell No.:	131A
Synonym:	Terbufos
P.C. Code:	105001
DP Barcode:	D258987
Submission:	S564874
Action Requested:	Please write Terbufos Toxicology chapter.

COMMENT: Attached is the Toxicology chapter on Terbufos. The Terbufos toxicology database is complete. There are sufficient data for selecting acute and chronic dietary endpoints and short-term and intermediate dermal and inhalation endpoints for risk assessment. Chronic dermal and chronic inhalation exposure assessments are not required, based on the use pattern of Terbufos. There are no toxicology data gaps *per se*. However, **confirmatory data** are required for NTE data for the hen study.

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NOTE: Do to time constraints, neither EXECUTIVE SUMMARIES nor Amended DERs were generated for this chapter. The previous Toxicology Chapter [Hazard Assessment] was included in the HED Chapter of the RED for Terbufos [dated 9/2/94]. Since that time, an acute neurotoxicity study, a subchronic neurotoxicity study, a two-generation reproduction study, several subchronic dermal toxicity studies, and an acute inhalation study have been submitted and reviewed. Additionally, Terbufos has been evaluated/re-evaluated by the HED Hazard Identification Assessment Review Committee and FQPA Safety Factor Committee.

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TERBUFOS: TOXICOLOGY CHAPTER FOR RED

DP Barcode No. D 258987 Reregistration Case No. 0109
Submission No. S 564874 P.C. Code No.105001
Tox. Chem. No. 131A

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TOXICOLOGY EXECUTIVE SUMMARY

The Terbufos toxicology database is complete. There are sufficient data for selecting acute and chronic dietary endpoints and short-term and intermediate dermal and inhalation endpoints for risk assessment. Chronic dermal and chronic inhalation exposure assessments are not required, based on the use pattern of Terbufos.

Terbufos, S-[[(1,1-dimethylethyl)thio]methyl]O,O-diethyþhosphorodithioate[IUPAC}; S-(*t*T-butylthio)methyl O,O-diethyl-phosphorodithioate; S-tert-butylmercaptomethyl O,O-diethyl dithiophosphate, is an organophosphate insecticide/nematicide applied as a granular formulation by soil incorporation, during planting or post-emergence of terrestrial food and feed crops [bananas, corn, sorghum, and sugar beets; 40CFR §180.352].

Terbufos is a cholinesterase inhibitor, and it produces the associated clinical cholinergic signs, such as tremors, irregular gait, salivation, hindleg weakness in rats and dogs and decreased cholinesterase activity [RBC, plasma, brain] in rats and dogs following acute, subchronic, and chronic exposure *via* the oral, dermal, and inhalation routes of exposure.

In acute toxicity studies, Terbufos exhibits severe toxicity *via* the oral, dermal, and inhalation routes of exposure. Because of lethality, dermal and eye irritation could not be assessed, and the dermal sensitation study was waived.

Terbufos did not cause acute delayed neurotoxicity in hens, and there was no evidence of neuropathology in the acute, subchronic, and chronic toxicity studies in rats, in the subchronic and chronic dog studies, or the mouse long-term study. In the acute neurotoxicity study in rats, excessive salivation, miosis, ataxia, stupor, decreased forelimb grip strength, slightly impaired locomotion, and tremor were observed, but motor activity was comparable among the groups. Also, no treatment-related effects were observed on motor activity or in the functional observation battery parameters measured in the subchronic neurotoxicity studies in rats.

Terbufos did not produce developmental toxicity, and there was no evidence of malformations or decreases in the numbers of pups and/or litters or surviving offspring. There is no indication of an increased sensitivity of offspring in rats or rabbits after prenatal and/or postnatal exposure to Terbufos. Reduced male fertility was observed in the 2-generation reproduction study in rats.

Terbufos did not alter the spontaneous tumor profile in either the Long Evans rat or CD-1 mouse carcinogenicity study, and both studies were considered adequate by the HED RfD/Quality Assurance Peer Review Committee. Terbufos was negative in all of the required mutagenicity battery assays. Terbufos is classified as a Group E.

DATA GAPS

There are no data gaps *per se*. However, **confirmatory data** are required for NTE in the hen.

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1. TOXICOLOGY DATA BASE

The toxicological data base on Terbufos is **complete** and supports reregistration eligibility.

A. Acute Toxicity

The acute toxicity data indicate that Terbufos is acutely toxic by the oral [Toxicity Category I], dermal [Toxicity Category I], and inhalation [Toxicity Category I] routes. Terbufos is considered a severe eye and dermal irritant, although dermal and eye irritant assessments were not possible due to deaths of all rabbits within 24 hours of exposure. Terbufos did not produce delayed neurotoxicity in hens. In an acute neurotoxicity study in rats, Terbufos caused miosis, ataxia, decreased forelimb grip strength, slightly impaired locomotion, tremors, and inhibition of plasma, RBC, and brain cholinesterase at dose levels of 0.30 mg/kg and above. Table 1 summarizes the acute toxicity data for Terbufos.

Table 1. Acute Toxicity of Terbufos

Guideline No.	Study Type	MRIDs #	Results	Toxicity Category
870.1100/§81-1	Acute Oral - rat	00029863✘	LD_{50} = ≈1.5 mg/kg	I
870.1200/§81-2	Acute Dermal - rabbit	258710✘	LD_{50} = ♂ 0.81/♀ 0.93 combined 0.87mg/kg	I
870.1300/§81-3	☛Acute Inhalation - rat	41538101	LC_{50} = 1.7 µg/L	I
870.2400/§81-4	Primary Eye Irritation	00044957✘	all rabbits died	I♪
870.2500/§81-5	Primary Skin Irritation	00044957✘	all rabbits died	I♪
870.2600/§81-6	Dermal Sensitization	-	waived due to lethality	N/A
870.6100/§81-7	Delayed Neurotoxicity	00037472✘	not a delayed neurotoxicant	N/A
870.6200/§81-8	☛Acute Neurotoxicity	44672003	NOAEL = 0.15 mg/kg; LOAEL = 0.30 mg/kg, based on findings in FOB and ChEI	N/A

☛ were data gaps in the Reregistration Eligibility Document]; ✘ Accession No.; ♪ assume [could not be determined]; above studies satisfy the acute toxicity data requirements [OPPTS 870.1100, 870.1200, 870.1300, 870.2400, 870.2500, 870.6100, 870.6200; formerly §81-1 through §81-5, §81-7, §81-8] for Terbufos. No dermal sensitization study was performed due to lethality.

B. Subchronic Toxicity

Available studies are adequate to satisfy subchronic testing requirements for Terbufos. **The subchronic data base provides evidence that Terbufos has anticholinesteraseactivity in rats and dogs, and clinical signs [tremors, irregular gait, decreased activity, salivation, muscle weakness] have been observed in rats following subchronic exposures. Decreased plasma cholinesterase activity was observed following oral exposure to rats and dogs, and in rats exposed dermally to a formulation of Terbufos, tremors and cholinesterase**

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inhibition [plasma, RBC, brain] were observed.

Subchronic toxicity in rats

In a subchronic [3 month] toxicity study (MRID 00109446), Terbufos [90% a.i.] was administered *via* the diet to groups of 20 Sprague-Dawley rats/sex/group at doses of 0, 0.125, 0.25, 0.5, and 1 ppm [0, 0.00625, 0.0125, 0.025, and 0.050 mg/kg/day, respectively].

There were no treatment-related deaths or clinical signs. Body weights and body-weight gains were comparable among the groups for both sexes, and there were no adverse effects noted in food consumption. No treatment-related effects were observed in the hematology, clinical chemistry [except cholinesterase activity], ophthalmoscopic, or urinalysis parameters measured. Plasma cholinesterase activity was decreased throughout the study in both sexes at the high-dose level [males 26%-42%/females 36%-52%]. There was no apparent treatment-related effect on either RBC or brain cholinesterase activity in either sex. There were no treatment-related differences in organ weight, macroscopic, or microscopic findings in either sex, with the exception of an increase in the incidence of mandibular lymph node hyperplasia in both sexes at the high dose compared to the controls and an increase in the incidence of mesenteric lymph node hyperplasia in the high-dose females [lower dose levels were not examined].

The NOAEL for cholinesterase inhibition is 0.25 ppm [0.0125 mg/kg/day], and the LOAEL for cholinesterase inhibition is 0.5 ppm [0.025 mg/kg/day], based on plasma cholinesterase inhibition in both sexes. The systemic NOAEL is 0.25 ppm [0.0125 mg/kg/day], and the LOAEL is 0.5 ppm [0.025 mg/kg/day], based on increased liver weight in females and an increase in liver extramedullary hematopoiesis in both sexes.

In a subchronic inhalation study [MRID 00258710], groups of 10 Sprague-Dawley rats/sex/group were exposed to vapors of technical Counter Terbufos for 3 weeks [8 hours/day, 5 days/week] at target concentrations of 0, 0.005, 0.01, 0.05, and 0.10 mg/m^3 and observed for an additional 2 weeks [Accession No. 258710]. During the first week of exposure, the mean analytical concentrations ranged from 2 to 44 percent of target concentrations. Because of these low values, the rats were exposed for an additional 2 weeks for a total of 3 weeks. The mean analytical concentrations of test material for weeks 2 and 3 were 0.0117, 0.0243, 0.0458, and 0.0946 mg/m^3 for males and 0.0112, 0.0256, 0.0468, and 0.1001 mg/m^3 for females. Chamber concentrations were not well controlled, and wide variations in the daily concentration were noted.

Two females that were exposed to 0.1001 mg/m^3 died during week 3 following signs of cholinesterase inhibition. At the conclusion of the exposure phase, brain cholinesterase was significantly lower for males and females exposed to a target concentration of 0.1 mg/m^3. Histopathological evaluations were not performed. Although the mean concentrations for the males in the two highest dose groups were 0.0458 mg/m^3 and 0.094 mg/m^3, respectively, the lowest daily mean chamber concentrations were selected as the values for the NOAEL and LOAEL.

The NOAEL is 0.0098 mg/m^3 [0.01 μg/L] **and the LOAEL is 0.0394 mg/m^3** [0.04 μg/L], **based on brain cholinesterase inhibition.**

28-day oral dog study

In a 28-day oral study in dogs [MRID 40374701] performed specifically to define the plasma cholinesterase

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activity NOAEL, plasma cholinesterase inhibition was observed at dose levels of 2.5 µg/kg/day [14%-23%] and above. Tthe NOAEL for plasma cholinesterase activity was determined to be 5 µg/kg/day.

Dermal toxicity in rats

Executive Summary: In a repeated 28-day dermal toxicity study (MRID 44690501), female Sprague Dawley rats (10 rats/dose) were treated dermally [10%-15% of total body surface] with an unpulverized 20 CD formulation of Terbufos (20.65% a.i.) as neat material moistened with saline at doses of 0, 5 and 10 mg/kg/day for 6 hours/day, 5 days a week for 4 weeks. Sites were covered with a gauze bandage. In the range-finding study [MRID 44450601], Sprague-Dawley rats [3/sex/group] were treated dermally as above with Terbufos CR formulation [20.72% a.i.] at dose levels of 0, 10, 25, 50, and 125 mg/kg/day.

In the **main study**, no mortality was observed and there were no biologically-significant or treatment-related effects on clinical signs, body weight, body-weight gain, or food consumption. There were no effects observed on plasma, RBC, or brain cholinesterase activity at any time point. No dermal irritation was observed. In the **range-finding study**, two of the three females at the highest dose level were found dead [days 12 and 13]. All other rats survived. Males at the highest dose level displayed a decreased body-weight gain at day 26 only compared to the controls. Body-weight gains were comparable among the groups at all other dose levels [both sexes]. Signs of cholinergic toxicity [tremors, excessive salivation, irregular gait, labored breathing, anogenital staining] were observed occasionally in both sexes at the highest dose level [day 9 on]. Plasma cholinesterase activity was depressed at the three highest dose levels in both sexes [19%-62%, 40%-74%, and 58%-96% with increasing dose, respectively] at all time points measured. RBC cholinesterase activity was depressed in both sexes at the three highest dose levels throughout the study [34%-73%, 56%-86%, and 81%-98% with increasing dose, respectively], but only on day 26 in females at 10 mg/kg/day. Brain cholinesterase activity was depressed at the highest dose level in both sexes [males 54%/females 78%], and both sexes at the next two dose levels displayed non-significant decreases [12%-25%] in brain cholinesterase activity.

The NOAEL is 10 mg/kg/day [2 mg/kg/day a.i.], based on plasma cholinesterase inhibition in both sexes at 25 mg/kg/day (LOAEL).

This guideline 21-day dermal toxicity study is classified Acceptable, and it satisfies the guideline requirement [§82-2; 870.3200] for a 21-day dermal toxicity study.

Executive Summary: In a 28-day dermal toxicity study (MRID No. 44520501), AC 92100 15G (terbufos, 16.06% a.i.) was administered topically to the clipped dorsal region (intact skin) of Sprague Dawley [Crl:CD BR] rats (10/sex/dose). Animals received daily dose of 0, 2, 5, 10 or 25 mg AC 92100 15G/kg/day (0, 0.32, 0.8, 1.6 or 4.0 mg a.i./kg/day) for 6 hours per day, 5 days per week, for 4 weeks. Animals were weighed before study initiation, weekly and at study termination. Plasma and erythrocyte cholinesterase determinations were performed on days 1, 5, 12, and 26 (at termination). At necropsy, the right halves of brains were analyzed for cholinesterase activity.

There were no statistically significant treatment-related effects on dermal reactions. No treatment-related effects on body weight, mortality, food consumption, hematology, blood chemistry, organ weights, gross or microscopic pathology were observed. Clinical signs related to cholinesterase inhibition such as localized fine tremors in the forepaws and hindlegs were seen during weeks 3 and 4 in females treated at 25 mg/kg/day.

There was a statistical significant reduction in plasma cholinesterase activity in males treated at 5 mg/kg/day

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or above (ranged from 17% to 60%) and in both males and females treated at 10 or 25 mg/kg/day (38% to 93%).

There was a statistical significant reduction in erythrocyte cholinesterase activity in both males and females treated at 10 or 25 mg/kg/day (ranged from 31% to 96%).

There was a statistical significant reduction in brain cholinesterase activity in males treated at 5, 10 or 25 mg/kg/day and in females treated at 10 or 25 mg/kg/day (ranged from 18% to 70%).

No dermal irritation was observed in this study; the NOAEL for dermal irritation is equal to or greater than 25 mg/kg/day (4 mg/kg/day a.i., HDT). The cholinesterase LOAEL is 5 mg/kg/day (0.8 mg/kg/day a.i.) based on statistically significant inhibition of plasma cholinesterase (ranged from 17.2% to 18.6% relative to controls) and brain cholinesterase activity (8.9%). The cholinesterase NOAEL is 2 mg/kg/day (0.32 mg/kg/day a.i.).

This guideline 21-day dermal toxicity study is classified Acceptable, and it satisfies the guideline requirement [§82-2; 870.3200] for a 21-day dermal toxicity study.

C. Chronic Toxicity and Carcinogenicity

Available studies are adequate to satisfy chronic toxicity and carcinogenicity testing requirements for Terbufos. The only treatment-related effect observed in dogs following chronic exposure to Terbufos was cholinesterase inhibition. In the rat, there was no increase in tumor incidence following Terbufos exposure *via* the diet for two years at dose levels that were considered adequate, based on cholinesterase activity [plasma, RBC and brain] inhibition in both sexes. In mice, there was no increase in tumor incidence in either sex at dose levels that were considered adequate, based on decreased body-weight gain.

The carcinogenicity issue has been discussed by the Health Effects Division-RfD/Quality Assurance Peer Review Committee [HED Document No. 011197, 7/17/93]. The Committee considered both the rat and mouse long-term studies to be adequate and classified Terbufos as a **Group E**.

Chronic toxicity in dogs

Executive Summary: In a 1-year oral toxicity study (Accession No. 00263678), male and female Beagle dogs (6 per sex/group) were given Terbufos (89.6%) *via* corn oil capsules at concentrations of 0, 15, 60, 90 [initially 240 until week 8], and 120 [initially 480 until week 7] µg/kg/day. Control dogs [8/sex] received corn oil capsules.

Treatment-related deaths occurred at the initial mid-high [one female] and initial high-dose [one male and three females] levels. Behavioral changes associated with cholinesterase inhibition [tremors, listless ness, excessive salivation, weak hind legs, diarrhea/red-tinged feces] were also observed in both sexes at these two dose levels [one male and three females at the mid-high dose; three males and 6 females at the high dose]. Following the reduction in dose levels, the overt signs of cholinesterase inhibition disappeared. Both sexes at the high-dose level lost weight during the first 7 weeks prior to the reduction in dose level. Males [65% of control] and females [57% of control] at the mid-high dose level displayed lower body-weight gains during the same time interval. Thereafter, both sexes and dose levels displayed a greater body-weight gain than the controls. Plasma cholinesterase activity was consistently depressed at all dose levels in both sexes throughout the study [≈40% at low, >60% all other dose levels]. RBC cholinesterase activity was depressed throughout the study in both sexes at the two highest dose

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levels only. With respect to brain cholinesterase activity, there was a dose-related decrease in cerebral cholinesterase activity in the males but comparable values were observed in cerebellar cholinesterase activity. In the females, there was no dose response in cerebral activity but cerebellar cholinesterase activity was decreased at the two highest dose levels [dose-related].

In a 28-day oral study in dogs [MRID 40374701] performed specifically to define the plasma cholinesterase activity NOAEL, plasma cholinesterase inhibition was observed at dose levels of 2.5 µg/kg/day [14%-23%] and above. The TES Committee determined that the NOAEL for plasma cholinesterase activity was 5 µg/kg/day.

The NOAEL for plasma cholinesterase activity is 5 µg/kg/day.

This guideline [OPPTS 870.4100/§83-1] chronic toxicity study [including the 28-day study] in the dog study is classified **Acceptable**, and it satisfies the guideline requirement for a chronic feeding study in non-rodents.

Chronic toxicity/carcinogenicity in rats

In a one-year dietary toxicity study [MRID 40098602], 30 CD COBS rats/sex/group were feed Terbufos [89.6% a.i.] at dose levels of 0 [corn oil], 0.125, 0.5, and 1 ppm [males 0.007, 0.028, and 0.055 mg/kg/day; females 0.009, 0.036, and 0.071 mg/kg/day] for one year.

There was no adverse effect on mortality, body weight, body-weight gain, or food consumption in either sex, and clinical signs were comparable among the groups.

Decreased plasma cholinesterase activity was observed in the high-dose males at 6 [25%] and 12 [29%] months and in the high-dose females throughout the study (at 1.5 [42%], 3 [44%], 6 [51%], and 12 [33%] months). RBC cholinesterase activity was comparable among thegroups in both sexes throughout the study. Brain cholinesterase activity was decreased in both sexes [males 8%/females 10%] at the high-dose level at study termination.

Testes weight was decreased in the high-dose males, and kidney weight was decreased in the mid- and high-dose females [not dose-related], but there were no microscopic findings in either organ.

The systemic NOAEL is 1 ppm [males 0.055/females 0.071 mg/kg/day], the highest dose tested. The NOAEL for cholinesterase inhibition is 0.5 ppm [males 0.028/females 0.036 mg/kg/day], based on plasma and brain cholinesterase activity inhibition in both sexes.

In a combined chronic toxicity/carcinogenicity study in rats [MRID 00049236], Terbufos [89.6% a.i.] was administered to 55 Long Evans rats/sex/dose via the diet at dose levels of 0 ppm, 0.25 ppm [0.0125 mg/kg/day], 1 ppm [0.05 mg/kg/day], and 2 ppm [0.1 mg/kg/day] for two years. The high-dose was raised to 4 ppm [0.2 mg/kg/day] after 6 weeks, then to 8 ppm [0.4 mg/kg/day] at 12 weeks (females lowered to 0.2 mg/kg/day at 16 weeks] for the remainder of the study.

There were an increase in mortality in both sexes [males 24%/females 27%] at the high-dose level compared to the controls, and males in the mid- [19%] and high-[10%] dose groups also displayed an increase in mortality. At the 8 ppm dose level, the high-dose females displayed tremors, hyperactivity, and excessive salivation. Body weight and body-weight gains were decreased in both sexes at the high-dose level compared to the control throughout the study [NOTE: The DER is old and does not contain details; insufficient time precluded a more detailed EXECUTIVE SUMMARY]. Food consumption was decreased at the high-dose level for both sexes.

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There were no treatment-related findings in the hematology, clinical pathology [except cholinesterase inhibition] or urinalysis parameters measured in either sex. There were no treatment-related macroscopic or microscopic findings in either sex. There was no treatment-related increase in tumor incidence following oral exposure to Terbufos for two years at dose levels considered adequate but not excessive in either sex compared to the controls.

Females at the 1 ppm dose level displayed decreased RBC cholinesterase activity [43%] and brain cholinesterase activity [20%]. Females at the higher dose level displayed greater brain the RBC cholinesterase inhibition. At the lowest dose level, RBC cholinesterase activity was decreased [males 12%/females 15%] compared to the controls.

The NOAEL for systemic toxicity was not attained in this study, based n increased mortality in the males. The NOAEL for cholinesterase inhibition is <0.25 ppm [<0.0125 mg/kg/day], based on RBC cholinesterase inhibition observed at all dose levels.

This guideline [§83-5; OPPTS 870.4300] chronic toxicity/carcinogenicity study in the rat is classified **Acceptable,** when considered along with the 1-year study, and it satisfies the guideline requirement for a chronic toxicity/carcinogenicity study in the rodent.

Carcinogenicity in mice

In a carcinogenicity/toxicity study in mice [MRID 40089603], Terbufos [89.6% a.i.] was administered to 65 Charles River CD-1 mice/sex/group at dose levels of 0, 3 ppm [0.45 mg/kg/day], 6 ppm [0.9 mg/kg/day], and 12 ppm [1.8 mg/kg/day] for 18 months.

There were a greater number of mice dying on test at the high-dose level in both sexes [males 13%, 9%, 6%, and 27%; females 27%, 15%, 22%, and 35%] than in the control and other treatment groups. Clinical signs were comparable among the groups for both sexes. Body weight was significantly lower in both sexes at the high-dose level throughout the study [males 94%/females 89% of control at sacrifice]. During the first week, the high-dose males did not gain weight, and the high-dose females lost weight, suggesting an initial palatibility problem. Overall body-weight gain was lower in both sexes [males 90%/females 80% of control] at the high-dose level compared to the control. A slight decrease in food consumption was noted in the high-dose females only. There were no apparent treatment-related changes in the hematology parameters measured in either sex. There were no treatment-related effects on organ weights, and gross or microscopic findings were comparable among the groups in both sexes at both the interim and final sacrifice. There was no increase in the incidence of spontaneous tumors in either sex. NOTE: No cholinesterase measurements were performed.

The systemic NOAEL is 6 ppm [0.9 mg/kg/day], and the systemic LOAEL is 12 ppm [1.8 mg/kg/day], based on a slight increase in mortality and a reduction in body-weight gain in both sexes.

This guideline [OPPTS 870.4200/§83-5] carcinogenicity study in mice is classified **Acceptable,** and it satisfies the guideline requirement.

D. Developmental Toxicity

Available developmental toxicity studies are adequate to satisfy guideline requirements. Terbufos was not a developmental toxicant in either the rat or rabbit. A weight-of-evidence analysis of the toxicity data base for Terbufos indicates there is no concern for quantitative or qualitative increased susceptibility to rats following pre-/or postnatal exposure in the developmental and reproductive toxicity studies. No evidence of enhanced

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susceptibility was observed for Terbufos following *in utero* exposure to pregnant rabbits. There was no evidence of effects being produced in fetuses at lower doses as compared to maternal rabbits nor was there evidence of an increase in severity of effects at or below maternally toxic doses.

Developmental toxicity in rats

In a developmental toxicity study (MRID 00147533/258787),25 COBS(CD) female rats/dose were administered Terbufos [87.8% a.i.] *via* gavage at doses of 0, 0.05, 0.10 and 0.2 mg/kg/day from day 6 to 15 of gestation.

There was no frank maternal toxicity. No treatment-related deaths and no clinical signs of toxicity were observed, and body weights, body-weight gains, and food consumption were comparable among the groups. Pregnancy rates were comparable among the groups, there were no abortions, and there were comparable numbers of corpora lutea and implantations among the groups. There was a slight increase in the litter incidence of resorptions, and the number of litters with two or more resorptions was higher at the high-dose level than in the control. Post-implantation loss was increased at the high-dose level compared to the control, and it exceeded the historical control incidence [1.3 *vs* 1.2]. The number of fetuses/litter was slightly lower [91% of control] than the control at the mid- and high-dose levels. Fetal body weight was comparable among the groups, and there was no evidence of a developmental effect at any treatment level, based on visceral, external and skeletal examinations of the fetuses.

In a range-finding study, 5 COBS(CD) female rats/dose were administered Terbufos [87.8% a.i.] *via* gavage at doses of 0, 0.4, 0.8, 1.4, 3.0, and 6.0 mg/kg/day from day 6 to 15 of gestation. An additional two groups [0.05 and 0.2 mg/kg/day] of 5 females/group were similarly dosed in a second study following the deaths of those in the higher dose groups.

Due to a protocol deviation, the dose levels in the first study were twice those specified in the protocol. At the three highest dose levels, all dams were dead on day 7. Dams at the 0.8 mg/kg/day dose level died during gestation days 8 and 9, and those at 0.4 mg/kg/day died during days 10-16 of gestation. At the 0.4 mg/kg/day dose level, all dams displayed either complete or significant early resorptions.

There was no evidence of maternal or fetal toxicity and no deaths at the 0.05 and 0.2 mg/kg/day dose levels. Pregnancy rates were comparable among the groups.The incidence of early resorptions in the control, 0.05 and 0.2 mg/kg/day dams was 1.0, 1.7, and 0.6, respectively. Based on these findings, the dose levels of 0.05, 0.1, and 0.2 mg/kg/day were chosen for the definitive study.

The maternal NOAEL is ≥0.2 mg/kg/day, the highest dose tested.

The developmental NOAEL 0.1 mg/kg/day, and the developmental LOAEL is 0.2 mg/kg/day, based on increases in early fetal resorptions, the number of litters with two or more resorptions, and postimplantation losses.

This guideline [OPPTS 870.3700/§83-3(a)] developmental toxicity study in the rat is classified **Acceptable**, and it satisfies the guideline requirement for a developmental toxicity study in rats.

Developmental toxicity in rabbits

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In a developmental toxicity study [MRID 40886301/40966401], Terbufos (89.6% a.i.) was administered to pregnant female New Zealand White rabbits (17/dose) by gavage at dose levels of 0 (corn oil), 0.05, 0.10,0.25, and 0.5 mg/kg body weight/day from days 7 through 19 of gestation.

All rabbits survived until study termination. One mid- [day 22] and one high-dose [day 21] does aborted. There was an increased incidence of soft/liquid feces at the high-dose level. Body weight was comparable among the groups, but body-weight gains were lower at the two highest dose levels [0.16 kg] compared to the control [0.26 kg] during the dosing period. Food consumption was slightly lower during days 20-29 at the high-dose level compared to the control. There were no effects on pregnancy rats, the number of corpora lutea, the number of implantations, litter size, live/dead fetuses, and uterine weights. There were a greater number of does with resorptions at the high-dose level compared to the control and other dose groups [19%, 42%, 38%, 31%, and 71% with increasing dose]. Fetal body weights were slightly lower [≈95% of control] at the high-dose level compared to the control. There were no apparent effects on gross, soft tissue, or skeletal parameters in the fetuses.

The maternal systemic NOAEL is 0.10 mg/kg/day, and the maternal LOAEL is 0.25 mg/kg/day, based on decreased body-weight gain during dosing. The developmental toxicity NOAEL is 0.25 mg/kg/day, and the developmental toxicity LOAEL is 0.5 mg/kg/day, based on decreased fetal body weight and increased percent of does with resorptions.

This guideline [OPPTS 870.3700/§83-3(b)] developmental toxicity study is classified **Acceptable**, and it satisfies the guideline requirement for a developmental toxicity study in the rabbit.

E. Reproductive Toxicity

Available data are adequate to satisfy the guideline requirements for reproductive toxicity testing. Reproductive effects, as evidenced by decreased fertility and decreased pregnancy rate were observed following exposure to Terbufos. No evidence of enhanced susceptibility was observed for Terbufos following pre- and/or postnatal exposure in the two-generation reproduction study in rats.

Reproductive toxicity in rats

In a 2-generation reproduction study [MRID 43649402] in Sprague-Dawley derived COBS CD rats [25/sex/dose], Terbufos [89.6%] was administered at dietary levels of 0, 0.5, 1.0, and 2.5 ppm [males 0.0350-0.0372, 0.0704-0.0742, and 0.1795-0.1943 mg/kg/day/females 0.0430-0.0436, 0.0854-0.0885, and 0.2188-0.2396 mg/kg/day, respectively] for 9 weeks prior to mating [males and females], as well as during gestation and lactation. There were two litters per generation. NOTE: There was an additional group administered 5 ppm, but this group was terminated at week 6 due to excessive mortality of the females [6 of 25].

With the above-noted exception, there was no apparent, treatment-related mortality in either sex. The only clinical sign observed among the groups was a dose-related increase in soft stools in the P1 females at week 28. Body weight/gains were comparable among the males in both generations. The high-dose females displayed a negative body-weight gain during lactation [F1a, F1b, F2a, F2b]. Food consumption was not adversely affected.

There was a dose-related decrease in cholinesterase activity [plasma, RBC, and brain] in both sexes at both time points measured. Females displayed the greater effect in both generations. Plasma cholinesterase activity was significantly inhibited at the mid-dose in P1 females [61%] and F1 rats [males 20%/females 5-%] and at the high-dose in both sexes [males P1 46%/F1 53%; females P1 94%/F1 87%] of both generations. RBC cholinesterase

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activity was decreased slightly [11%-15%] in both sexes of both generations, mainly at the high-dose level. Brain cholinesterase activity was inhibited in both sexes and both generations at the mid- [males P1 8%/F1 8%; females P1 22%/F1 21%] and high-dose [males P1 29%/F1 34%; females P1 66%/F1 59%] levels and in the low-dose P1 females [7%].

At the high-dose level, male fertility and the number of pregnant females were decreased during the second mating in both generations [P1-F1b 79%/80%; F1-F2b 61%/63%] compared to the conrol and first matings. \\\No adverse effects were observed in the following parameters: live birth index, sex ratio, viability index in the F_0 generation [both litters], or day 0 pup weight. At the high-dose level, there was a decrease in the number of pups per litter only at day 21 *post partum* in the first litter of the F_0 generation but at all time points in the second litter of the F_0 generation and both litters of the F_1 generation. The lactation index of both litters of both generations were significantly decreased compared to the controls. The viability index was significantly decreased at the high-dose level in the second litter of the F_1 generation. Pup body weights were significantly decreased compared to the controls from day 4 *post partum* on [all litters].

The NOAEL for cholinesterase inhibition [plasma, RBC, brain] is 0.5 ppm [males 0.0350-0.0372/females 0.0430-0.0436 mg/kg/day]. The LOAEL for cholinesteraseinhibition [plasma, RBC, brain] is 1.0 ppm [males 0.0704-0.0742/females 0.0854-0.0885 mg/kg/day]. The reproductive/developmental NOAEL is 1.0 ppm [males 0.0704-0.0742/ females 0.0854-0.0885 mg/kg/day], and the reproductive/developmental LOAEL is 2.5 ppm [males 0.1795-0.1943/females0.2188-0.2396 mg/kg/day], based on decreases in the pregnancy rate and male fertility. The maternal toxicity/offspringNOAEL is 1.0 ppm [males 0.0704-0.0742/females0.0854-0.0885 mg/kg/day], and the maternal toxicity/offspring LOAEL is 2.5 ppm [males 0.1795-0.1943/females 0.2188-0.2396 mg/kg/day], based on decreased body-weight gain in females during lactation and lower pup body weights during lactation days 14 and 21.

This guideline [§83-4; OPPTS 870.3800] 2-generation reproduction study in the rat is classified **Acceptable**, and it satisfies the guideline requirement for a 2-generation reproduction study in rats.

F. Mutagenicity

Available mutagenicity studies are adequate to satisfy the guideline requirements. **Terbufos is not mutagenic.**

Terbufos was negative with and without microsomal activation in *Salmonella typhimurium* assay [TA 100, TA 1535, TA 1537, TA 98, E. Coli, WP-2-uvrA], reflected a negative mutagenic potential in DNA damage/repair assays, did not increase chromosomal aberrations, and was negative in the CHO/HGPRT assay. In a dominant lethal test in rats , there was an apparent compound-related effect at the highest dose tested [numbers of viable implants were slightly but significantly reduced at mating 9]. Additionally, implant efficiency was significantly lower at mating 7, as well as suggestively, but not significantly, lower during matings 8, 9, and 10.

All Subdivision F guideline mutagenicity requirements have been satisfied.

G. Metabolism

Available metabolism data are adequate to satisfy the guideline requirements. Following oral administration, Terbufos was rapidly and extensively absorbed from the gastrointestinal tract, and the metabolites were rapidly eliminated. There is no evidence of bioaccumulation.

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The absorption, distribution, metabolism, and excretion of Terbufos were studied [MRID 2348801] in groups of male and female CD rats administered a single oral dose of radiolabeled Terbufos *via* gavage at dose levels of 0.1 or 0.4 mg/kg or a 14-day repeated oral dose of 0.1 mg/kg/day of unlabeled Terbufos followed by a single oral dose of 0.1 mg/kg radiolabeled Terbufos.

Following oral administration, Terbufos is rapidly and extensively absorbed in the gastrointestinal tract, metabolized, and the metabolites [not parent compound] are rapidly excreted, mainly *via* the urine, within the first 24 hours after dosing. There is no evidence of bioaccumulation. The predominant radiolabeled compound found in the feces was Terbufos. Neither Terbufos nor its metabolites accumulated in any tissue to any extent, but the highest level of radiolabel was found in the lungs. The percent of the administered dose detected in the urine [168 hours] ranged from 69.3% to 86.3%, in feces ranged from 5.4% to 17.4%, and in expired air ranged from 2.6% to 4.2%. No sex differences were observed. Following repeat exposure, there appeared to be a shift towards greater urinary elimination. The proposed metabolism of Terbufos is *via* desulfuration and/or sulfoxidation, followed by hydrolysis of the phosphorus-sulfur bond and exzymatic S-methylation. The proposed final step is S-oxidation.

This guideline [OPPTS 870.7485/§85-1] metabolism study is classified **Acceptable**, and it satisfies the guideline requirement for a metabolism study.

H. Neurotoxicity

Available neurotoxicity studies are adequate to satisfy the guideline requirements. Terbufos was not a delayed neurotoxicant in the hen study. Treatment-related neuropathology was not observed in any available toxicity study on Terbufos.

Acute neurotoxicity in hens

In an acute delayed neurotoxicity study [MRID 00037472], hens were given a single oral dose of Terbufos [86.5%] at a dose of 40 mg/kg by oral gavage and observed for 21/22 days. NOTE: No DER was generated for this study previously, and due to time constraints, a DER/EXECUTIVE SUMMARY was not generated here.

No delayed neurotoxicity was seen in hens given a single oral dose of Terbufos at 40 mg/kg. No neuropathology was observed. This study did not assess for the potential of Terbufos to inhibit neurotoxic esterase [NTE] in hens.

This guideline [OPPTS 870.6100/§81-7] acute oral study in the hen is classified **Acceptable**, and it satisfies the guideline requirement for a delayed neurotoxicity study in the hen. **Data on NTE are required as confirmatory data.**

Acute neurotoxicity in rats

In an acute oral neurotoxicity study (MRID No. 44672003) 5 groups of 7 week old Sprague-Dawley CD® rats (20/sex/group) were given a single oral dose (by gavage) of Terbufos, 89.7% a.i., in corn oil at doses of 0, 0.15, 0.30, and 0.90 mg/kg of body weight. Before the initiation of the main study, an additional 10 females were exposed to 0.90 mg/kg of Terbufos to determine survivability (satellite study).

Clinical signs such as anogenital stains, oral stains, lethargy, reduced defecation and food consumption were observed in females treated with 0.90 mg/kg of the test substance (main study and satellite study). A red exudate

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from the eye was reported in one male treated at the 0.30 mg/kg dose level. These symptoms did not persist beyond day 5 of the study. One female treated at the 0.90 mg/kg dose level died 5.5 hours after treatment with the test article. No other mortalities were reported during the test period.

The first indication of a compound-related effect (miosis) detected by the FOB was reported at the 0.3 mg/kg and 0.90 mg/kg doses approximately 6 hours after exposure to the test substance in both males and females. In addition to miosis, females at the 0.90 mg/kg dose level showed evidence of excessive salivation, lacrimation, ataxia, stupor, soiled coat, decreased forelimb strength, slightly impaired locomotion, and tremors. These symptoms resolved within the first week after treatment with the test article and were no longer evident at the Day 7 observation period.

Ten animals/sex/dose were tested for plasma, erythrocyte, and brain cholinesterase activity (PChE, RChE, and BChE, respectively). Significant cholinesterase inhibition was reported in both males and females at the 0.30 mg/kg and 0.90 mg/kg doses.

Gross necropsy examination and histopathology testing of animals that were euthanized at the end of the study revealed no compound-related abnormalities.

Under the conditions of this study, **the NOAEL for acute neurotoxicity and plasma cholinesterase inhibition is 0.15 mg/kg/day. Given the findings of the FOB testing and cholinesterase inhibition, the LOAEL is established at 0.30 mg/kg/day.**

This guideline [OPPTS 870.6200/§81-8] acute neurotoxicity study in rats is classified **Acceptable**, and it satisfies the guideline requirement for an acute neurotoxicity study in rats.

Subchronic neurotoxicity in rats

In a subchronic neurotoxicity study (MRID 44842302), Terbufos [AC 92100] (89.7% a.i.) was administered in the diet to 20 Crl:CD (SD) IGS BR rats/sex/dose at dose levels of **0 ppm, 0.5 ppm, 0.8 ppm, or 5.0 ppm [males]/3.0 ppm [females]** for at least 85 days [10 rats/sex/cholinesterase group] or 13 weeks [10 rats/sex/neurobehavioral group]. These dose levels corresponded to **0.036, 0.059, and 0.369 mg/kg/day, respectively, in males; 0.042, 0.064, and 0.251 mg/kg/day, respectively, in females.**

No adverse or treatment-related effects were observed on mortality or clinical signs. The high-dose males displayed slightly decreased body weights [94%-97% of control] throughout the study compared to the controls, but the high-dose females displayed body weights that were comparable to or greater than the controls throughout the study. Body-weight gains were decreased [86%-93% of control] in the high-dose males throughout the study, although the magnitude of the deficit diminished with time. There was no adverse effect on body-weight gain in the females. Food consumption was comparable to/greater than the control in the treated groups [both sexes].

There were no treatment-related effects observed on ophthalmoscopy, and motor activity was comparable among the groups for both sexes throughout the study. There were no significant and treatment-related differences relative to the controls in any of the parameters monitored in the functional observational battery in either sex.

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There was a dose-related inhibition of plasma cholinesterase activity [males ≈70%/females ≈90% inhibition] and RBC acetylcholinesterase activity [≈100% inhibition] throughout the study in both sexes. At study termination, brain cholinesterase activity [males 55%-58%/females 68%-71% inhibition] was decreased at the high-dose level in both sexes, with the magnitude of the inhibition greater in the females than in the males.

No treatment-related macroscopic and microscopic lesions were observed in either sex. Brain weight data were not provided.

The NOAEL for systemic toxicity is 0.8 ppm [0.059 mg/kg/day], and the LOAEL for systemic toxicity is 5 ppm [0.369 mg/kg/day], based on decreased body weight and body-weight gains in males. No neurobehavioral or neuropathological effects were observed in either sex.

The NOAEL for inhibition of plasma cholinesterase and RBC acetylcholinesterase activity [both sexes] is 0.5 ppm [0.036 (males)/0.042 (females) mg/kg/day] and the LOAEL for inhibition of plasma cholinesterase and RBC acetylcholinesterase activity [both sexes] is 0.8 ppm [0.059 (males)/0.064 (females) mg/kg/day].

The NOAEL for brain cholinesterase activity is 0.8 ppm [0.059 (males)/0.064 (females) mg/kg/day] and the LOAEL for brain cholinesterase activity is 5 (males)/3 (females) ppm [0.369 (males)/0.251 (females) mg/kg/day].

This guideline [OPPTS 870.6200/§81-6] subchronic neurotoxicity study in rats is classified **Acceptable**, and it satisfies the guideline requirement for a subchronic neurotoxicity study in rats

I. Dermal absorption

There is no adequate dermal absorption study. A 100% absorption factor should be used for Terbufos.

J. Reference Dose [RfD] for Chronic Oral Exposure

$$\text{Reference Dose: } \textbf{Chronic RfD: } \frac{0.005 \text{ mg/kg/day}}{100} = 0.00005 \text{ mg/kg/day}$$

K. Uncertainty Factor/FQPA Considerations

The following evaluation of the chemical Terbufos is provided to address FQPA considerations on the sensitivity of infants and children. The FQPA Safety Factor Committee recommended that the **10X FQPA safety factor** should be **removed** [FQPA Safety Factor Committee report dated 8/6/99, HED Doc. No. 013616] due to the fact that no increased susceptibility was found in offspring, and there is a complete toxicological data base [*Memorandum*: B. Tarplee to W. Hazel dated 8/2/99; HED Doc. No. 013598].

L. Recommendation for a Developmental Neurotoxicity Study

The developmental neurotoxicity study in the rat is **not required**. Confirmatory NTE data for the hen study is required.

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2. TOXICITY END-POINT SELECTION

The Hazard Identification Assessment Review Committee [HIARC] met on several occasions to evaluated the entire toxicological database on Terbufos and selected the relevant toxicity endpoints, taking into consideration the use pattern and exposure information on this chemical. The selected toxicological endpoints and the doses for risk assessment are summarized in the table below. The details are present in the HIARC report, dated August 17, 1999 [HED Document No.], which is appended.

SUMMARY OF TOXICOLOGY ENDPOINT SELECTION

EXPOSURE SCENARIO	DOSE (mg/kg/day)	ENDPOINT	STUDY
Acute Dietary	NOAEL= 0.15 UF = 500√	Plasma ChE inhibition in both sexes	Acute Oral Neurotoxicity in Rats
		Acute RfD = 0.0003 mg/kg	
Chronic Dietary	NOAEL=0.005 UF = 100	Plasma ChE Inhibition in both male and female dogs	Chronic/28-day Toxicity -Dog
		Chronic RfD = 0.00005 mg/kg/day	
Short- and Intermediate-Term (Dermal) ***20 CR only***	NOAEL= 2	plasma, RBC, brain ChE Inhibition observed at higher doses in range-finding study	28-Day Dermal Study on 20 CR formulation in Female Rats
Short- and Intermediate-Term (Dermal) ***15G only***	NOAEL = 0.32	Plasma and brain ChE Inhibition	28-Day Dermal Study on 15G formulation in Rats
Long-Term Dermal	This risk assessment is not required.		
Short- and Intermediate-Term [Inhalation]	NOAEL= 0.00001 mg/L	Plasma , RBC, brain ChE Inhibition	Subchronic Inhalation Study in Rats
Long-Term Inhalation	This risk assessment is not required.		

√ [10 for intraspecies; 10 for interspecies; 5 for species sensitivity (dog *vs* rat)];
* MOE for worker exposure risk assessments = 100; no registered residential uses

3. DATA GAPS

There are no data gaps *per se*. However, **confirmatory data** are required. NTE data for the hen should be submitted.

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