

US EPA ARCHIVE DOCUMENT



CASE GS0109 TERBUFOS PH 04/15/82

CHEM 105001 Terbufos (S-(((1,1-dimethylethyl)thio)

BRANCH EEB OISC 40 TOPIC 05054543

FORMULATION 00 - ACTIVE INGREDIENT

FICHF/MASTER ID 00087718 CONTENT CAT 01

Roberts, S.; Wineholt, R.L. (1976) Static 96-hour Toxicity Study of Terbufos in Bluegill Sunfish and Brown Trout; Laboratory No. 6E-3166. (Unpublished study received Nov 24, 1976 under 2749-427; prepared by Cannon Laboratories, Inc., submitted by Aceto Chemical Co., Inc., Flushing, N.Y.; COL1226951-A)

SUBST. CLASS = S.

DIRECT RVW TIME = 6 hrs. (MH) START-DATE 10/4/82 END DATE 11/2/82

REVIEWED BY: James D. Felkel
TITLE: Wildlife Biologist
ORG: Ecological Effects Branch, Hazard Evaluation Division (TS-769)
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SIGNATURE: *James D. Felkel* DATE: 12/8/82

APPROVED BY:
TITLE:
ORG:
LOC/TEL:

SIGNATURE: DATE:

DATA EVALUATION RECORD

1. Chemical: Terbufos (Shaughnessy No. 105001)
2. Formulation: Technical, 86% a.i. (F. Betz 6/30/79 review)
3. Citation: Roberts, S. and Wineholt, R. 1976. Static 96-hour toxicity study of terbufos in bluegill sunfish and brown trout. Unpublished study by Cannon Laboratories, Inc. for Aceto Agricultural Chemical Corp. (MRID No. 00087718)
4. Reviewed By: James D. Felkel, Wildlife Biologist
Ecological Effects Branch
Hazard Evaluation Division (TS-769)
5. Date Reviewed: November 1, 1982
6. Test Type: Freshwater fish acute LC₅₀ (static bioassay)
 - A. Test Species: Bluegill sunfish (Lepomis macrochirus)
Brown trout (Salmo trutta)
7. Reported Results: The bluegill LC₅₀ is 0.77 (0.71-0.83) ppb and the brown trout LC₅₀ is 16 (8-31) ppb.
8. Reviewer's Conclusions:

These studies are considered scientifically sound and indicate that terbufos is very highly toxic to both test species with LC₅₀ values as follows: bluegill, 0.77 (0.72-0.83) ppb; brown trout, 20 (12.6-34.3) ppb. These studies meet the intent of proposed guidelines (7/10/78) for this test type.

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METHODS

Bluegill sunfish (Lepomis macrochirus, representing a warm water species) and brown trout (Salmo trutta, representing a cold water species) of between 35 and 75 mm in length and 0.5 to 3.0 grams in weight were used. Care was taken when selecting the fish in each group to insure that the largest fish in the group was never more than one and one-half times the size of the smallest fish in the group. Bluegills were obtained from Kurtz's fish Hatchery, Elverson, Pa. and trout were obtained from Greenwalk Trout Hatchery, Bangor, Pa.

Stock fish were acclimatized for 10 days prior to bioassay under dynamic flow conditions with continuous aeration and charcoal filtration of the water. The water temperature was kept at 19 + 2°C and 15 + 2°C for the bluegills and trout, respectively. The fish received a standard commercial fish food (Purina Trout Chow #2) daily until 2 days prior to testing at which time feeding was discontinued. Only fish from stock groups with a mortality rate of less than 10 percent, 48 hours prior to testing, were chosen.

An initial range finding experiment was conducted using 5 fish per concentration level. The range was found by beginning at 1 ppm and decreasing the amount of the test material by a factor of 10 until a range between no deaths and 100 percent deaths had been established.

Once the general toxic level of the test material was determined, a logarithmic series of 5 concentrations were selected for toxicity determination. Twenty fish per concentration level were used. The test material was dispensed into the bioassay vessels in an acetone solution. Acetone concentrations were 100 ppm or less at all dose levels. An acetone control at 100 ppm was included as a vehicle test. No deaths occurred in the acetone control.

The bioassay vessels used in this experiment were 20-liter all glass aquaria containing 10 liters of well water. the quality of the water is expressed in the following table:

Turbidity	0.	NTU		
Color	0.		pH	7.5
Odor	None		Conductivity (mmhos/cm)	520.
Total Hardness	14.7	GPG	Est Tds by conductivity	320. PPM
Iron (Fe)	0.7	PPM	Chemical Oxygen Demand	PPM
Manganese (Mn)	0.0	PPM	Compensated Hardness*	16. GPG
			Nitrate (N)	1.7 PPM
			Silica	7.0 PPM
Calcium	8.7	GPG	- Anions-	
Magnesium	6.0	GPG	Chloride	0.6 GPG
Sodium	0.5	GPG	Sulfate	2.8 GPG
Potassium	0.1	GPG	Nitrate	0.4 GPG
			Hydroxide Alk.	GPG
			Carbonate Alk.	GPG
			Bicarbonate Alk.	11.5 GPG
Total Cations	15.3	GPG	Total Anions	15.3 GPG

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The test water was kept at $19 \pm 2^\circ\text{C}$ and $15 \pm 2^\circ\text{C}$ for sunfish and trout, respectively. During the test, continuous aeration was discontinued to prevent loss of the material. However, if the dissolved oxygen dropped below 4.0 ppm for sunfish or 5.0 ppm for trout, intermittent aeration was used to maintain the necessary levels.

The fish were observed for 96 hours. The number of fish surviving, general behavior, dissolved oxygen level and pH were recorded for each concentration level at 6, 24, 48, 72 and 96 hours.

RESULTS

Bluegill sunfish, exposed to "Terbufos" at concentrations levels of 0.00037 and 0.00021 ppm, exhibited no observable signs of abnormal behavior. At concentration levels between 0.0010 and 0.00065 ppm, bluegill sunfish exhibited pectoral fin erection, partial and total loss of equilibrium and death (see Table 3).

At concentration levels between 0.075 ppm and 0.001 ppm, brown trout exhibited partial loss of equilibrium and death (see Table 4).

No abnormal behavior or high level of mortality was observed in the untreated control and vehicle control groups (see Table 3 & 4).

Survival rates, dissolved oxygen count and pH for "Terbufos" are presented in Table 1 for bluegill sunfish and in Table 2 for brown trout.

LC₅₀ determinations were calculated according to Litchfield, J.T., Jr. and Wilcoxon, F., "A Simplified Method of Evaluating Dose-Effect Experiments", J. Pharm. and Exp. Therap. 96, 99-113 (1949).

The calculated results are presented below with the LC_x value listed first and the confidence limits at P = .05 following in parenthesis.

Bluegill Sunfish

LC1 = 0.00056 ppm (0.00056 ppm to 0.00056 ppm)
LC50 = 0.00077 ppm (0.00071 ppm to 0.00083 ppm)
LC99 = 0.0011 ppm (0.0011 ppm to 0.0011 ppm)

Brown Trout

LC1 = 0.0002 ppm (0.0001 ppm to 0.0004 ppm)
LC50 = 0.016 ppm (0.008 ppm to 0.031 ppm)
LC99 = 0.80 ppm (0.41 ppm to 1.55 ppm)

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REVIEWER'S EVALUATION

These studies were previously reviewed by F. Betz of EEB (reviews attached) on 6/30/79. Methods reported were generally consistent with proposed guidelines (7/10/78). The intermittent aeration was considered acceptable by F. Betz because of the low volatility of Terbufos, the apparently infrequent use of the aeration to maintain dissolved oxygen levels, and the fact that results indicate a similar or greater toxicity than is seen in other acute freshwater fish studies (and thus the aeration did not appear to reduce exposure by volatilizing the test material).

Specific loading information was not provided. It was reported that fish weighted from 0.5 - 3.0 grams and that 20 fish were tested per 20 liter vessel. If fish averaged more than 0.8 g each for each of the vessels (16 g of fish/vessel), loading would exceed accepted protocols (Stephan, 1975). Reduced loading, rather than aeration, should be used to avoid low dissolved oxygen levels. Other discrepancies from Stephan (1975) include a shorter acclimation/holding period (10 days vs. 14) and food-withholding period (48 vs. 96 hours) than specified.

EPA computer analysis (attached) indicates an LC_{50} of 0.77 (0.72 - 0.83) ppb for the bluegill and 20 (12.6 - 34.3) ppb for the brown trout. Thus, Terbufos is considered very highly toxic to these species.

CONCLUSIONS

1. Category: Core
2. Rationale: Discrepancies from accepted protocols are not considered severe enough to prevent studies from meeting the intent of proposed guidelines (7/10/78).
3. Repairability: N/A

TABLE 1
SURVIVAL, DISSOLVED OXYGEN AND pH DATA FOR BLUEGILL SUNFISH

CONCENTRATION (PPH)	SURVIVORS AT (HOURS)						PERCENT SURVIVAL	DISSOLVED OXYGEN (PPH) AND pH AT (HOURS)								
	6	24	48	72	96			6	24	48	72	96				
	00	pH	00	pH	00	pH		00	pH	00	pH	00	pH			
0.0010	20	10	8	6	1	5	2.8	7.6	5.0	7.6	4.0	8.0	5.0	8.0	4.4	8.0
0.00075	20	20	20	14	12	60	2.6	7.6	5.0	7.6	4.2	8.0	5.0	7.6	4.2	8.0
0.00065	20	20	19	17	17	85	2.4	7.6	4.6	7.6	5.2	8.0	5.2	8.0	4.7	8.0
0.00037	20	20	20	20	20	100	2.8	7.6	4.3	7.4	4.0	8.0	6.2	7.4	5.6	8.0
0.00021	20	20	20	20	20	100	2.6	7.8	4.0	7.6	3.8	8.0	5.7	8.0	4.8	8.0
Untreated Control	20	20	20	20	20	100	2.8	8.0	5.0	7.6	4.0	8.0	5.0	8.0	4.6	8.0

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TABLE 2
SURVIVAL, DISSOLVED OXYGEN AND PH DATA FOR BROWN TROUT

CONCENTRATION (PPH)	SURVIVORS AT (HOURS)				PERCENT SURVIVAL	DISSOLVED OXYGEN (PPH) AND PH AT (HOURS)												
	6	24	48	72		96	6	24	48	72	96	DO	PH	DO	PH	DO	PH	DO
0.075	16	4	2	2	2	10	5.3	7.4	5.4	7.6	5.6	7.6	5.3	7.2	5.3	7.2	5.3	7.2
0.037	20	20	20	0	0	40	5.4	7.5	5.3	7.4	5.7	7.4	5.2	7.3	5.3	7.3	5.3	7.4
0.01	20	20	15	15	15	75	5.6	7.4	5.5	7.6	5.7	7.6	5.2	7.6	5.4	7.6	5.4	7.6
0.0032	20	20	20	19	19	95	5.8	7.6	5.7	7.4	5.7	7.6	5.0	7.6	5.3	7.6	5.3	7.6
0.001	20	20	20	18	18	90	5.6	7.4	5.6	7.6	5.6	7.6	5.2	7.2	5.2	7.2	5.2	7.6
Untreated Control	20	20	19	19	19	95	5.7	7.5	5.7	7.5	5.6	7.6	5.3	7.6	5.4	7.6	5.4	7.6

TABLE 3
BEHAVIORAL OBSERVATIONS OF BLUEGILL SUNFISH

CONCENTRATION (ppm)	NUMBER OF FISH	6	24	48	72	96
0.0010	20	N(20)	N(8)O(10)PE,PLE(2)	D(2)PE,PLE(6) N(2)	N(1)O(2)PE,PLE(5)	O(5)PE,PLE(1)
0.00075	20	N(20)	N(19)PE,PLE(1)	N(14)PE,PLE(6)	N(8)PE,PLE(6)D(6)	N(8)O(2)PE, PLE(4)
0.00065	20	N(20)	N(20)	N(19)O(1)	N(17)D(2)	N(17)
0.00037	20	N(20)	N(20)	N(20)	N(20)	N(20)
0.00021	20	N(20)	N(20)	N(20)	N(20)	N(20)
Untreated Control	20	N(20)	N(20)	N(20)	N(20)	N(20)

O = DEATH
N = NORMAL
PE = PECTORAL FIN ERECTION
PLE = PARTIAL LOSS OF EQUILIBRIUM
() = NUMBER OF FISH AFFECTED

N. VILLIOTTI - (1971)

TABLE 4
BEHAVIORAL OBSERVATIONS OF BROWN TROUT

CONCENTRATION (ppm)	NUMBER OF FISH	6	24	48	72	96
0.075	20	M(16)D(4)	M(3)D(12)PLE(1)	M(2)D(2)	M(2)	M(2)
0.037	20	M(20)	M(20)	M(20)	M(8)D(12)	M(8)
0.01	20	M(20)	M(20)	M(15)D(5)	M(15)	M(15)
0.0032	20	M(20)	M(20)	M(20)	M(19)D(1)	M(19)
0.001	20	M(20)	M(20)	M(20)	M(18)D(2)	M(18)
Untreated Control	20	M(20)	M(20)	M(19)D(1)	M(19)	M(19)

0 = DEATH
M = NORMAL
PE = PECTORAL FIN ERECTION
PLE = PARTIAL LOSS OF EQUILIBRIUM
() = NUMBER OF FISH AFFECTED

FELKEL TERBUFOS BLUEGILL LC50 (00087718)

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
1	20	19	95	0.002002716
0.75	20	8	40	25.17223
0.65	20	3	15	0.1288414
0.37	20	0	0	9.536743E-05
0.21	20	0	0	9.536743E-05

THE BINOMIAL TEST SHOWS THAT 0.65 AND 1 CAN BE USED AS STATISTICALLY SOUND CONSERVATIVE 95 PERCENT CONFIDENCE LIMITS, BECAUSE THE ACTUAL CONFIDENCE LEVEL ASSOCIATED WITH THESE LIMITS IS GREATER THAN 95 PERCENT.

AN APPROXIMATE LC50 FOR THIS SET OF OATA IS 0.7844547

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
2	0.1198138	0.7813182	0.7298097	0.8375477

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
7	0.1759924	1	0.9947893

SLOPE = 14.35865
 95 PERCENT CONFIOENCE LIMITS = 8.334989 ANO 20.38231

LC50 = 0.7742599
 95 PERCENT CONFIDENCE LIMITS = 0.7258437 AND 0.8343169

LC10 = 0.6315945
 95 PERCENT CONFIDENCE LIMITS = 0.5420606 AND 0.6814715

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ABBOTT'S CORRECTION CANNOT BE USED WITH THIS DATA SET.

FELKEL TERBUFOS BROWN TROUT LC50 (00087718)

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB.(PERCENT)
75	20	18	90	0.02012253
37	20	12	60	25.17223
10	20	5	25	2.069473
3.2	20	1	5	0.002002716
1	20	2	10	0.02012253

THE BINOMIAL TEST SHOWS THAT 10 AND 75 CAN BE USED AS STATISTICALLY SOUND CONSERVATIVE 95 PERCENT CONFIDENCE LIMITS, BECAUSE THE ACTUAL CONFIDENCE LEVEL ASSOCIATED WITH THESE LIMITS IS GREATER THAN 95 PERCENT.

AN APPROXIMATE LC50 FOR THIS SET OF DATA IS 25.70182

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS
4	0.08365949	19.75374	12.61142 34.67664

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
5	0.1218952	1	0.07649469

SLOPE = 1.4707
95 PERCENT CONFIDENCE LIMITS = 0.9572274 AND 1.984173

LC50 = 20.07667
95 PERCENT CONFIDENCE LIMITS = 12.57963 AND 34.31935

LC10 = 2.748891
95 PERCENT CONFIDENCE LIMITS = 0.9233062 AND 5.047164

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TEST: Fish Acute LC₅₀

SPECIES: Bluegill Sunfish

RESULTS: 96 Hour LC₅₀ = 0.77 (0.71-0.83) ppb

No observed effect at 0.37 ppb and below. Lowest level at which mortality occurred was 0.65 ppb. Abnormal behavior (pectoral fin erection, partial loss of equilibrium) occurred at 0.75 and 1.0 ppb.

Additional Information:

Water was intermittently aerated, whenever D.O. fell below 4.0 ppm. Based upon D.O. values reported at 6, 24, 48, 72 and 96 hours, it appears that some aeration was required. Water temperature was 19° ± 2° and loading factor was 0.05-0.3 gm/l, both of which are acceptable.

CHEMICAL: Terbufos (86% a.i.)

TITLE: Static 96-Hour Toxicity Study of Terbufos in Bluegill Sunfish
and Brown Trout

AUTHOR: Cannon Labs. Inc.

STUDY DATE: October 21, 1976

ACCESSION NO. 226951

REGISTRANT: Aceto Agricultural Chemicals Corp.

VALIDATION CATEGORY: Core

CATEGORY REPAIRABILITY:

This study is classified core despite the fact that the water was intermittently aerated because: (1) aeration was not continuous, (2) the parent material and its degradates are not highly volatile in water (according to Bob Carsel, EFB), (3) the vapor pressure of terbufos is 2.6×10^{-4} mm hg. at 25°C which suggests relatively low volatility (according to Bob Carsel, EFB) and (4) other Bluegill LC₅₀ studies conducted under static and flow thru conditions gave higher LC₅₀ values.

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TEST: Fish Acute LC₅₀

SPECIES: Brown Trout (Salmo trutta)

RESULTS: 96 Hour LC₅₀ = 16 (8-31) ppb.

Five percent mortality occurred in the control group and some mortality (5-90%) occurred at all test levels.

Abnormal behavior such as pectoral fin erection or partial loss of equilibrium occurred only at the highest test level (75 ppb).

ADDITIONAL INFORMATION:

Water was intermittently aerated whenever D.O. fell below 5.0 ppm. All reported D.O. values are greater than 5.0 ppm, thus the actual amount of aeration is unknown. Water temperature was 15° ± 2°C and loading factor was 0.05 to 0.3 g/l.

CHEMICAL: Terbufos (86% a.i.)

TITLE: Static 96-Hour Toxicity Study of Terbufos in Bluegill Sunfish and Brown Trout.

AUTHOR: Cannon Labs. Inc.

STUDY DATE: October 21, 1976

ACCESSION NO. 226951

REGISTRANT: Aceto Agricultural Chemicals Corp.

VALIDATION CATEGORY: Core

CATEGORY REPAIRABILITY:

This study is classified core despite the fact that the water was intermittently aerated because: (1) aeration was not continuous, (2) according to Bob Carsel, EFB, the parent material and its degradates are not highly volatile in water and based on its vapor pressure (2.6×10^{-4} mm hg @ at 25°C), terbufos is not highly volatile and (3) other Rainbown trout LC₅₀ studies conducted by other researchers yielded similar results.

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