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DATES

(TCR038)

DATA EVALUATION RECORD

PAGE 1 OF

CASE GS0109 TERBUFOS 04/15/82 CHEM 105001 Terbufos (S=(((1,1=dimethylethyl)thio) BRANCH EEB OISC 40 TOPIC 05054543 FORMULATION OG - 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 (Unpublished study received Nov 24, 1976 under 2749-427; prepared by Cannon Laboratories, Inc., submitted by Aceto Chemical Co., Inc., Flushing, N.Y.; CDL:226951-A) SUBST. CLASS = S. DIRECT RYW TIME = 6 hrs. (MH) START-DATE 10/4/82 REVIEWED BY: James D. Felkel TITLE: Wildlife Biologist ORG: Ecological Effects Branch, Hazard Evaluation Division (TS-769) LOC/TEL: Crystal Mall #2, Room 1112, 703-557-3113 SIGNATURES ene d. That DATE: 12/8/82 APPROVED BY: TITLE: ORG: LOC/TEL:

SIGNATURE:

DATA EVALUATION RECORD

- 1. Chemical: Terbufos (Shaughnessy No. 105001)
- 2. Formulation: Technical, 86% a.i. (F. Betz 6/30/79 review)
- 3. <u>Citatition:</u> 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 Unpublished study by Cannon Laboratories, Inc. for Aceto Agricultural Chemical Corp.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. <u>Test Species</u>: Bluegill sunfish (<u>Lepomis macrochirus</u>)
 Brown trout (<u>Salmo trutta</u>)
- 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 $\rm IC_{50}$ 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.

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 form 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 kep at $19 \pm 2^{\circ}\text{C}$ and $15 \pm 2^{\circ}\text{C}$ 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 conentrations 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 litters of well water. the quality of the water is expressed in the following table:

Turbidity	0.	NTU	
Color	0.	pH Conductivity (mmhos/cm)	7.5 520.
Odor Total Hardness Iron (Fe) Marganese (Mn) —Cation— Calcium Magnesium Sodium Potassium	0.7 1 0.0 1 8.7 0 6.0 0 0.5 0	Est Tds by conductivity Chemical Oxygen Demand GPG Compensated Hardness* PPM Nitrate (N) PPM Silica - Anions- GPG Chloride GPG Sulfate GPG Nitrate GPG Hydroxide Alk. Carbonate Alk. Bicarbonate Alk.	320. PPM PPM 16. GPG 1.7 PPM 7.0 PPM 0.6 GPG 2.8 GPG 0.4 GPG GPG GPG GPG 11.5 GPG
Total Cations	15.3 G	GPG Total Anions	15.3 GPG

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

LCl = 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

LCl = 0.0002 ppm (0.0001 ppm to 0.0004 ppm) $LC_{50} = 0.016$ ppm (0.008 ppm to 0.031 ppm) $LC_{99} = 0.80$ 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 (Stephen, 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 foodwithholding 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 form accepted protocols are not considered severe enough to prevent studies from meeting the intent of proposed guidelines (7/10/78).
- 3. Repairability: N/A

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0.001	50,	70	50	œ)	82	8	5.6	5.6 7.4 5.6 7.6 5.6 7.6 5.2 7.2	5.6	7.6	5.6	7.6	5.3	7.2	5.2 7.6	7.
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TABLE 3 BENAVIORAL OBSERVATIONS OF BLUEGILL SUNFISH	, 96 0(5)PE,PLE N(8)0(2)PE PLE(4) N(17) N(20)	72 H(1)0(2)PE,PLE(5) H(6)PE,PLE(6)D(6) H(17)D(2) H(20)	1L SUNFISH 48 N(2) 0(2)PE,PLE(6) N(14)PE,PLE(6) N(19)0(1) N(20)	TABLE 3 SERVATIONS OF BLUEGI 24 N(B) O(10) PE, PLE(2) N(19) PE, PLE(1) N(20) N(20)	BENAVIORAL OB: (20) (20) (20) (20)	HUMBER OF F1SH 20 20 20 20	CDNCENTRATION (PPm) 0.0010 0.00075 0.00065
HUMBER OF FISH 20 N(20) N(8)0(10)PE,PLE(2) 20 N(20) N(19)PE,PLE(1) N(14)PE,PLE(6) 20 N(20) N(20) N(19) 0(1) 20 N(20) N(20) N(20) N(20)	N(20)	N(20)	N(20)	. N(20)	N (20)	20	0.0002 l
NUMBER OF FISH 6 24 48 N(2) 20 N(20) N(8)0(10)PE,PLE(2) D(2)PE,PLE(6) N(20) N(19)PE,PLE(1) N(14)PE,PLE(6) N(20) N(20) N(19)0(1)	N(20)	N (20)	H(20)			20	0.00037
NUMBER OF FISH 6 24 48 N(2) N(20) N(8)O(10)PE,PLE(2) D(2)PE,PLE(6) N(20) N(19)PE,PLE(1) N(14)PE,PLE(6)	(LON .	M(17)0(2)	(1)0(61)N.	N(20)	N(20)	20	0,00065
HUMBER OF 54 48 72 N(2) N(2) N(2) N(20) N(20) N(8)0(10)PE,PLE(2) D(2)PE,PLE(6) N(1)0(2)PE,PLE(5) 0(5)P	N(8)0(2)PE PLE(4)	N(&)PE,PLE(6)D(6)	N(14)PE,PLE(6)	N(19)PE,PLE(1)	N(20)	02	6.00075
FISH 6 24 48 72	0(5)PE,PLE	N(1)0(2)PE,PLE(5)	N(2) D(2)PE,PLE(6)	N(8)0(10)PE.PLE(2)	N(20)	20	0.0010
	81	77	87		' اه	NUMBER OF	CONCENTRATION (PPm)

O - DEATN N - HORMAL

7

PE - PECTORAL FIN ERECTION

PLE - PARTIAL LOSS OF EQUILIBRIUM () - NUMBER OF FISH AFFECTED

ð	TROUT
	BROWN
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TABLE 4	OBSERVATIONS OF
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श्र	N(2)	· (8)	N(15)	(61)×	N(18)	H(19)
77	#(2)	N(8)0(12)	N(15)	M(19)0(1)	H(18)D(2)	K(19)
85 -F	H(2)0(2)	H(20)	N(15)0(5)	H(20)	H(20)	H(19)0(E)
42	N(3)0(12)PLE(1)	N(20)	N(20)	H(20)	H(20)	N(20)
اه	H(16)0(4)	H(20)	H(20)	N(20)	N(20)	H(20)
NUMBER OF	20	20,	50	50	- 00	- 50
CONCENTRATION (PPm)	0.075	0.037	0.01	0.0032	0.001	Untreated Control

OFATH .

N - NORMAL

PE - PECTORAL FIN ENECTION PLE - PARTIAL LOSS OF EQUILIBRIUM

⁻ NUMBER OF FISH AFFEETED

FELKEL TERBUFOS BLUEGILL LC50 (00087718)

^^^^	**********	*********	·***********	********
CONC.	NUM8ER Exposed	NUMBE R DE AD	PERCENT DE AD	8 I NOM I AL
1 0.75	20	19	95	PROB.(PERCENT) 0.002002716
0.65	20 20	8 3	40 15	25.17223 0.1288414
0.37 0.21	20	Ŏ	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 CONFIGENCE LIMITS = 8.334989 ANO 20.38231

LC50 = 0.7742599 95 PERCENT CONFIDENCE LIMITS = 0.7258437 AND 0.8343169

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

FELKEL TERBUFOS BROWN TROUT LC50 (00087718)

*****	*************	*******	*******	*******
CONC.	NUMBER Exposed	NUMBE R OE AD	PERCENT OEAD	8 I NOM I AL
75 27	20	18	90	PROB.(PERCENT) 0.02012253
37 10	20 20	12	60	25.17223
3.2	20	5 1	25 5	2.069473 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 OATA IS 25.70182

RESULTS CALCULATEO USING THE MOVING AVERAGE METHOO
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 GOOONESS OF FIT PROBABILITY

5 0.1218952 1 0.07649469

SLOPE = 1.4707 95 PERCENT CONFIGENCE LIMITS = 0.9572274 ANO 1.984173

LC50 = 20.07667 95 PERCENT CONFIGENCE LIMITS = 12.57963 AND 34.31935

 TEST: Fish Acute LC50

SPECIES: Bluegill Sunfish

RESULTS: 96 Hour $LC_{50} = 0.77 (0.71-0.83)$ 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, whenver 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 $19^{\circ} \pm 2^{\circ}$ 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.

CATEGORY REPAIRABILITY:

VALIDATION CATEGORY: Core

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 x 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.

TEST: Fish Acute LC₅₀

SPECIES: Brown Trout (Salmo trutta)

RESULTS: 96 Hour $LC_{50} = 16 (8-31)$ 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 whenver 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 $15^{\circ} \pm 2^{\circ}$ C and loading factor was 0.05 to 0.3 q/1.

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 \times 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.