

8-8-88

DATA EVALUATION RECORD

PAGE 1 OF

CASE: GS0103

PHORATE FRSTR

CONT-CAT: 01

GUIDELINES: 72-1

MRID: 161822

Nicholson, R.; McNabb, T. (1986) Acute Toxicity of Thimet 20G to Rainbow Trout (*Salmo gairdneri*): Report #BW-86-6-2051; Study #451.1285.6108.103. Unpublished study prepared by Springborn Bionomics, Inc. 34 p. *4002/801*

REVIEW RESULTS:

VALID INVALID INCOMPLETE

GUIDELINE:

SATISFIED PARTIALLY SATISFIED NOT SATISFIED

DIRECT RVW TIME = *2 hr*

START DATE: *6/15/88*

END DATE: *6/15/88*

REVIEWED BY:

Ann Stavola

TITLE: *aquatic Biologist*

ORG: *HED/EEB*

LOC/TEL: *CM2 107 D 557 1354*

SIGNATURE: *Ann Stavola*

DATE: *6/15/88*

APPROVED BY:

Douglas J. Urban

TITLE: *Supervisory Biologist*

ORG: *HED/EEB*

LOC/TEL: *CM2 815H*

557-4365

SIGNATURE: *Douglas J. Urban*

DATE: *8/2/88*

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DATA EVALUATION RECORD

1. Chemical: Phorate
2. Test Material: Thimet 20G, 20% ai
3. Study Type: Freshwater Fish Static Acute

Species Tested: Rainbow Trout
(Salmo gairdneri)

4. Study ID: MRID No. 161822. Nicholson, R.; McNabb, T. (1986)
Acute Toxicity of Thimet 20G to Rainbow Trout
(Salmo gairdneri): Report No.: BW-86-6-2051,
Study No.: 451.1285.6108.103. Unpublished study
prepared by Springborn Bionomics, Inc.

5. Reviewed By: Ann Stavola
Aquatic Biologist
EEB/HED

Signature: *Ann Stavola*

Date: *18 July 88*

6. Approved By: Douglas Urban
Supervisory Biologist
EEB/HED

Signature: *Douglas Urban*

Date: *10/11/88*

7. Conclusion:

This study is scientifically sound and with a 96-hour LC₅₀ value of 45 (37 to 57) ug/L, Thimet 20G is very highly toxic to rainbow trout. This study meets EPA Guidelines requirements for an acute toxicity test on a coldwater fish with a granular formulation of phorate, up to and including 20 percent ai.

8. Recommendations: N/A

9. Background:

Formulated product testing of freshwater fish was required in the Phorate Registration Standard, 1983.

10. Discussion of Individual Tests: N/A

11. Material and Methods

- a. Test Animals - Species: Rainbow trout (Salmo gairdneri):
Total Length: mean = 47 mm; range = 39 to 53 mm; Wet
Weight: mean = 1.1 g; range = 0.63 to 1.38 g; Source:
a commercial supplier in California.
- b. Dosage - Nominal concentrations: 60, 36, 22, 13, 8,
4.8 ug/L as Thimet 20G. Also control and solvent control.
Mean measured concentrations: 11, 7.2, 4.3, 3.8, 1.6,
and 0.84 ug/L as phorate and converted to ug Thimet 20G
per liter. Dilution water: deionized reconstituted well
water, soft quality, pH 7.5. Stock solution: 0.06 g
Thimet 20G dissolved in 500 mL acetone; ultrasonicated
for 3 hours and supernatant used as stock solution.
- c. Study Design - Protocol closely follows "Standard
Practice for Conducting Acute Toxicity Tests with Fishes,
Macroinvertebrates and Amphibians (ASTM, 1980). The
test procedure was modified to provide complete solution
renewals at 48 hours of the exposure period. This was
done to maintain consistent exposure concentrations of
the test material. At 48 hours, all surviving fish were
transferred to a duplicate set of aquaria containing
freshly prepared solutions. Types of Test Chambers:
19.6-L glass aquaria containing 15 L of test solution.
Number of Organisms per Test Concentration: 10 rainbow
trout were placed in each aquarium. Loading Factor:
0.73 g biomass/L test solution; Photoperiod: 16L:8D;
Test Temperature: 13 °C.
- d. Statistical Analyses - The mean measured concentrations
(0 and 48 hours) tested and the corresponding mortality
data were used to estimate the LC₅₀ values and
95 percent confidence intervals with a computer program
(Stephan, 1982).

12. Reported Results:

The stability of Thimet 20G in freshwater was evaluated prior to conducting this study. The half-life of phorate was 97.7 hours. Therefore, a 48-hour static renewal procedure was used for this test.

Table 1 presents the concentrations measured throughout the study.

Table 1. Results of the analysis of the test solutions during the 96 hour static renewal exposure of rainbow trout (Salmo gairdneri) to Thimet 20G. Test solutions were renewed at 48 hours. Measured concentrations are based on the analyses for Phorate, the active ingredient (20%) of Thimet 20G.

Nominal Concentration (ug/L) As Thimet 20G	Measured Concentration as Phorate (ug/L)				Extrapolated Concentration as Thimet 20G (ug/L)
	0-Hour ^a	48-Hour ^a	96-Hour ^b	Mean (SD) ^c	
Control	< 0.32	< 0.24	< 0.25	< 0.32	< 1.6
Solvent Control	< 0.32	< 0.24	< 0.25	< 0.32	< 1.6
4.8	0.84	0.87	< 0.25	0.86 (0.021)	4.2
8.0	1.5	1.7	0.31	1.6 (0.099)	8.0
13	2.4	5.1	0.51	3.8 (1.9)	19
22	4.1	4.5	1.0	4.3 (0.28)	21
36	6.9	7.6	1.3	7.2 (0.50)	36
60	11	12	2.3	11 (1.1)	57

^aAnalysis of freshly prepared solutions.

^bAnalysis of 48-hour-old solutions.

^cMean (standard deviation) based on the analysis of freshly prepared solutions only.

Analysis of QA samples showed satisfactory analytical control with a recovery range of 105 to 108 percent.

Table 2 presents the cumulative mortality data and observations of symptomatic behavior in fish.

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Table 2. Concentrations tested and corresponding cumulative mortalities of rainbow trout (*Salmo gairdneri*) exposed to Thimet 20G for 24, 48, 72, and 96 hours. Concentrations are based on extrapolated mean measured Phorate concentrations.

Extrapolated Mean Measured Concentration (ug/L) as Thimet 20G	Cumulative Mortality (%)			
	24-Hour	48-Hour	72-Hour	96-Hour
Control	0	0	0	0
Solvent Control	0	0	0	0
4.2	0	0	0	0
8.0	0	0	0	0
19	0	0	0	0
21	0	0	0 ^a	0
36	0	0	0 ^a	20 ^b
57	0	0 ^c	20 ^{d,e}	80 ^f

^aOne of the surviving fish was lethargic.

^bSeveral of the surviving fish were at the surface of the test solution.

^cSeveral of the surviving fish exhibited a partial loss of equilibrium.

^dSeveral of the surviving fish exhibited a complete loss of equilibrium.

^eSeveral of the surviving fish were lethargic.

^fAll of the surviving fish were lethargic.

The calculated LC₅₀ values are:

Time	LC ₅₀ and 95% CI (ug/L)
24-hour	> 57
48-hour	> 57
72-hour	> 57
96-hour	45 (37 to 57)

Dissolved oxygen (DO) and pH were measured at 24-hour intervals for each test concentration. The pH values were 7.1 and 7.2 at 0-hour and 7.0 and 7.1 at 96 hours. DO was 9.4 to 9.6 ppm at 0-hour, but reduced to 2.5 to 4.9 ppm (24 to 42 percent saturation) at the end of the first 48-hour interval. The lowest level was recorded at 57 ug Thimet/L. At the end of 96-hour, DO levels were 3.9 to 5.7 ppm, and, again, the lowest level was at the highest test concentration. Temperature was maintained at 13 °C.

13. Study Authors' Conclusions/QA Measures:

The 96-hour LC₅₀ for rainbow trout exposed to Thimet 20G was 45 (37 to 57) ug/L. This indicates Thimet 20G is very highly toxic to rainbow trout. The NOEL was 21 ug/L.

QA Statement: "The data contained in this report were audited by the Quality Assurance Unit to assure compliance with the protocols, standard operating procedures and the pertinent EPA Good Laboratory Practice Regulations. . . ."

14. Reviewer's Discussion and Interpretation of Study:

- a. Test Procedures - The procedures were done in accordance with protocols recommended by EPA Guidelines and are acceptable.
- b. Statistical Analysis - The LC₅₀ value for 96-hours was recalculated with EEB's program, which is also based on a program developed by Stephan.
- c. Results and Discussion - EEB calculated the 96-hour LC₅₀ value to be 45 (37 to 57) ug/L. As their result agrees with ours, we accept their value as valid.

The data indicate that Thimet 20G is very highly toxic to rainbow trout.

d. Adequacy of Study

- 1) Classification - Core for granular phorate up to 20 percent ai.
- 2) Rationale - Scientifically sound study that followed EPA Guidelines. Formulated product testing was required.
- 3) Reparability - N/A

15. Completion of One-Liner for Study: N/A

16. CBI Appendix: N/A

STAVOLA PHORATE RAINBOW TROUT 06-14-88

96-hr

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
57	10	8	80	5.46875
36	10	2	20	5.46875
21	10	0	0	9.765625E-02
19	10	0	0	9.765625E-02
8	10	0	0	9.765625E-02
4.2	10	0	0	9.765625E-02

THE BINOMIAL TEST SHOWS THAT 0 AND +INFINITY 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 45.299

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
2	.2148829	45.299	37.02619	63.535

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
8	.4067773	1	.999881

SLOPE = 8.700694
95 PERCENT CONFIDENCE LIMITS = 3.15147 AND 14.24992

LC50 = 45.39221
95 PERCENT CONFIDENCE LIMITS = 37.1201 AND 57.11326

LC10 = 32.43516
95 PERCENT CONFIDENCE LIMITS = 17.49643 AND 39.01996
