DATA EVALUATION RECORD

1. **CHEMICAL:** Mon 13900, Shaughnessey Number: ~~999999~~ 111596

2. **TEST MATERIAL:** Mon 13900, 96.4% active ingredient, brown granules.

3. **STUDY TYPE:** A 96-hour static acute toxicity test with the Rainbow trout (Oncorhyncus mykiss).

4. **CITATION:** Bowman, Jane, and John Bucksath. 1991. Acute toxicity of Mon 13900 to Rainbow trout (Oncorhyncus mykiss) Guideline 72-1(c) Project ID ABC 39004 Submitted by Monsanto Agricultural Co. Performed by ABC Laboratories Inc., MRID No. 420197-41.

5. **REVIEWED BY:**

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Signature: Renée Lamb
Date: 6/17/93

6. **APPROVED BY:**

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7. **CONCLUSIONS:** This study appears to be scientifically sound and fulfills the data requirements for an acute toxicity test for freshwater fish. The 96 hour LC_{50} value for Rainbow trout (Oncorhyncus mykiss) exposed to Mon 13900 was 6.2 mg/L with 95% confidence limits of 4.9 and 7.8 mg/L. The NOEC was 1.2 mg/L. Therefore, Mon 13900 is classified as moderately toxic to coldwater fish.

8. **RECOMMENDATIONS:**
9. **BACKGROUND:** N/A
10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A
11. **MATERIALS AND METHODS:**

A. **TEST ANIMALS:** The fish, Rainbow trout (Oncorhyncus mykiss) were obtained from Mt. Lassen Trout Farms in Red Bluff, CA. They were received as eyed eggs. After hatching, they were maintained in a "living stream" (an enclosed system in which water is recirculated through the system) 4 weeks prior to testing. The fish were acclimated to approximate test temperatures in this system. There was a 16 hour light photoperiod with a 30-minute transition period.

The fish were acclimated to test temperature and hard

blended dilution water for 48 hours prior to testing. The fish were not fed during the acclimation and test periods. The trout has a mean wet weight of .79 ± .11 g and a mean standard length of 37 ± 2 mm. The biomass loading rate for the control fish was .47 g/L. Measurements were made on the control group at termination of the test.

B. **TEST SYSTEM:** The test chambers were five gallon glass vessels containing 15 liters of hard blended water which was equivalent to a depth of ≈ 29.9 cm.

The photoperiod in the room was maintained at 16 hours of light and 8 hours of darkness with a 30 minute transition period.

C. **DOSAGE:** The fish were exposed to 5 test concentrations and control. The test concentrations (nominal) used were 1.9, 3.8, 7.5, 15, and 30 mg of active ingredient per liter. These concentrations were chosen by range finding toxicity tests. Mean measured concentrations were 1.2, 2.4, 5.0, 7.6 and 20 mg a.i./L. A primary stock solution of Mon 13900 technical at a concentration of 9.96 mg/mL in acetone was used for subsequent dilutions.

D. **DESIGN:** Ten fish were randomly allocated to each test vessel within 30 minutes after the addition of test material. All vessels were monitored for mortality and sub lethal effects after 2.3 hours and once every 24 hours.

E. **STATISTICS:** The 96 hour LC_{50} value was determined by the statistical method of Stephan et al.

12. **REPORTED RESULTS:** Preliminary testing in hard blended water showed that Mon 13900 is not completely soluble, due to this, the recovery of Mon 13900 was expected to be low. However, the test material appeared to be stable in the system based on information supplied by the sponsor and the consistent measurements at 0 and 96 hours. The mean measured concentrations were 62 ± 7% of the nominal concentration.

Abnormal effects were noted in the 2.4, 5.0, 7.6, and 20 mg/L test concentrations. There was 100% mortality in the 20 mg/L concentration after 72 hours. There was partial mortality after 96 hours in the 5.0 and 7.6 mg/L concentrations. Table 5 presents the mortality and abnormal effects observed during the study.

A brown precipitate was on the bottom of all test chambers except the control chamber throughout the study. The amount of precipitate increased as the concentration of Mon 13900

increased.

Water quality data are presented an Table 6. DO ranged from 7.3 to 8.7 mg/L, 71 and 84% saturation at 12°C, respectively. The pH ranged from 7.9 to 8.6. The temperature ranged from ≈ 11.9 to 13.0°C on the continuous temperature recording for the first 72 hours of the study. However, a bad connection occurred between the probe and data logger resulting in a data logger recording of 14.4°C. The data logger was reading nearly the same temperature as the thermometer until the last day of the test. The last day of the test, the logger read 14.4 and the thermometer read 13.0°C.

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:**

The 96 hour LC_{50} value for Rainbow trout (Oncorhyncus mykiss) exposed to Mon 13900 was 6.2 mg/L with 95% confidence limits of 4.9 and 7.8 mg/L. The NOEC was 1.2 mg/L.

The study has a quality assurance statement signed by a quality assurance officer.

14. **REVIEWER'S DISCUSSION AND INTERPRETATION OF STUDY RESULTS:**

A. **TEST PROCEDURE:** This test is in accordance with EPA's SEP protocol, except there was no solvent control.

There was a brown precipitate in the test chambers, however, as the concentrations were measured, any effects from the presence of the precipitate are likely accounted for.

B. **STATISTICAL ANALYSIS:** The data was analyzed with EEB's Toxanal program using mean measured concentrations.

C. **DISCUSSION/RESULTS:** This study appears to be scientifically sound and fulfills the data requirements for an acute toxicity test for freshwater fish. The 96 hour LC_{50} value for Rainbow trout (Oncorhyncus mykiss) exposed to Mon 13900 was 6.2 mg/L with 95% confidence limits of 4.9 and 7.8 mg/L. The NOEC was 1.2 mg/L. Therefore, Mon 13900 is classified as moderately toxic to coldwater fish.

D. **ADEQUACY OF STUDY:**

(1) CLASSIFICATION: core
(2) RATIONALE: Although there was no solvent control, the pattern of mortality plus the relatively moderate toxicity of the chemical, indicate that no new information would be gained with a new study.
(3) REPARABILITY: N/A

lamb mon 13900 trout

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
20	10	10	100	9.765625E-02
7.6	10	8	80	5.46875
5	10	2	20	5.46875
2.4	10	0	0	9.765625E-02
1.2	10	0	0	9.765625E-02

THE BINOMIAL TEST SHOWS THAT 2.4 AND 20 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 6.164414

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
3	.1792363	6.344816	4.524058	9.146871

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
6	.5385812	1	.9999941

SLOPE = 9.271651
95 PERCENT CONFIDENCE LIMITS = 2.467363 AND 16.07594

LC50 = 6.164862
95 PERCENT CONFIDENCE LIMITS = 4.910118 AND 7.760902

LC10 = 4.497247
95 PERCENT CONFIDENCE LIMITS = 1.798424 AND 5.399984

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TABLE 2

Measured Concentrations from the Acute Toxicity Test
of MON 13900 to Rainbow Trout (*Oncorhynchus mykiss*)

Sample	Nominal Conc. (mg/L)	Measured Concentrations (mg/L)			Percent Nominal
		0-Hour	96-Hour	Mean	
Reagent Blank	---	<0.40	<0.40	<0.40	---
Control	---	<0.40	<0.40	<0.40	---
Level 1	1.9	1.1	1.2	1.2	63
Level 2	3.8	2.3	2.4	2.4	63
Level 3	7.5	5.2	4.8	5.0	67
Level 4	15	6.6	8.6	7.6	51
Level 5	30	19	21	20	67

Mean ±S.D. = 62 ±7%

TABLE 4

Acute Toxicity of MON 13900
to Rainbow Trout (*Oncorhynchus mykiss*)

	LC_{50} in milligrams/liter (ppm)				
Compound	2.3-Hour	24-Hour	48-Hour	72-Hour	96-Hour
MON 13900[a]	>20[b]	>20[b]	15[(1)]	8.6[(1)]	6.2[(2)]
			c	(5.0 and 20)[d]	(4.9 and 7.8)[d]

N = 10 fish per concentration

[a] Bioassay as conducted at 12°C (±1°C), mean fish weight and length, 0.70 (±0.11) g and 37 (±2) mm.

[b] Since no mortality occurred an LC_{50} could not be calculated.

[c] Insufficient mortality for the calculation of 95% confidence limits.

[d] 95% confidence limits.

The 96-hour no-observed effect concentration could be estimated at 1.2 mg/L, based on the lack of mortality and abnormal effects at this concentration.

LC_{50} calculated using:

(1) Binomial Method

(2) Probit Method

TABLE 5

Mortality and Behavioral Observations During the Acute Toxicity Test of MON 13900 to Rainbow Trout (*Oncorhynchus mykiss*)

Mean Measured Conc. mg/L	No. Placed in Test	2.3 Hours		24 Hours		48 Hours		72 Hours		96 Hours	
		Mort.	Observations	Cum. Mort.	Observations	Cum. Mort.	Observations	Cum. Mort.	Observations	Cum. Mort.	Observations
Control	10	0	10 N	0	10 N	0	10 N	0	10 N	0	10 N
1.2	10	0	10 N	0	10 N	0	10 N	0	10 N	0	10 N
2.4	10	0	10 N	0	10 N	0	10 N	0	7 N; 3 LR	0	1 OB/LR; 4 LR; 5 N
5.0	10	0	7 N; 3 LR/FG	0	8 LR; 2 Q/OB/LR	0	8 Q/LR; 2 DK/OB/LR/Q	0	7 LR/Q; 1 DK/LR/Q; 2 DK/OB/LR/Q	2	2 SUR/LR/DK; 2 SUR/DK/VO/LR; 4 DK/OB/LOE/LR
7.6	10	0	6 N; 2 OB/LR; 2 LR/FG	0	4 LR; 6 OB/LR/Q	0	3 LR; 1 SUR/LR; 4 OB/Q/LR; 2 DK/OB/LR/Q	4	3 DK/LR; 2 DK/OB/LR/Q; 1 DK/OB/LR/Q/LOE	8	2 OB/LOE/LR/Q
20	10	0	7 LR/FG; 1 OB/LR/FG; 2 SUR/LR/FG	0	9 OB/DK/Q/LR; 1 OB/LOE/DK/LR/Q	8	2 OB/LOE/LR/Q/DK	10	---	10	---

Key to Observations: N = Normal; LOE = Loss of Equilibrium; Q = Quiescent; SUR = Surfacing; OB = On Bottom of Test Vessel; DK = Dark Discoloration; LR = Labored Respiration; VO = Vertical Orientation; FG = Flared Gills

TABLE 6

Water Quality Measurements During the Acute Toxicity Test of MON 13900 to Rainbow Trout (*Oncorhynchus mykiss*)

Mean Measured Conc. (mg/L)	Water Quality														
	0-Hour			24-Hour			48-Hour			72-Hour			96-Hour		
	Temp.[a] °C	D.O.[b] mg/L	pH[c]	Temp. °C	D.O. mg/L	pH	Temp. °C	D.O. mg/L	pH	Temp. °C	D.O. mg/L	pH	Temp. °C	D.O. mg/L	pH
Control	12	8.6	8.3	12	7.9	8.1	12	7.8	8.0	12	7.9	8.0	12	8.0	8.0
1.2	12	8.7	8.6	12	8.0	8.4	12	7.8	8.1	12	7.9	8.2	12	8.0	8.1
2.4	12	8.5	8.6	12	8.0	8.4	12	7.7	8.2	12	7.5	8.1	12	7.5	8.0
5.0	12	8.5	8.6	12	7.9	8.1	12	7.6	8.2	12	7.3	8.0	12	7.4	7.9
7.6	12	8.5	8.5	12	7.8	8.3	12	7.7	8.2	12	7.5	8.2	12	7.5	8.1
20	12	8.5	8.6	12	7.7	8.4	12	7.5	8.3	12	7.3	8.4	12	7.6	8.3

[a] Temperature measured using a mercury thermometer

[b] Dissolved oxygen concentrations - Dissolved Oxygen Probe (YSI Model 54).

[c] pH - pH Probe (Corning Model 476182) used with a Corning Model 125 pH and mV meter.

NOTE: Dissolved oxygen saturation corrected for altitude at the test temperature of 12°C is 10.3 mg/L.