Data Evaluation

1. Chemical: Roundup (41.36% a.i.)

2. Citation: Surprenant, D.C., "Acute Toxicity of Roundup to the Water Flea (*Daphnia magna*," prepared by E.G. & G, Bionomics Aquatic Toxicology Laboratory, Wareham, Massachusetts, for Monsanto, St. Louis, Mo., Reg. No. 524-308, Submission 3/30/81.

3. Reviewed by: Miachel Rexrode
   Fishery Biologist
   Ecological Effects Branch, HED

4. Date Reviewed: July 6, 1981

5. Test Type: Aquatic Invertebrate LC_{50}

   A). Test Type: *Daphnia magna*

   B). Test material: Roundup (41.36% a.i.)
   (Isopropylamine salt of glyphosate)

   C). Report Result: The acute 48-hour LC_{50} of Roundup to *Daphnia magna* was estimated at 5.3 mg/l with confidence limits ranging from 4.4-6.3 mg/l.

6. Reviewers Conclusion: Data derived from this study appear to be scientifically sound and satisfy the regulatory requirments as stated in the EPA guidelines, 1978, for a formulation. The LC_{50} of 5.3 mg/l implies that Roundup formulation is moderately toxic to daphnids. The no discernable concentration level through 48-hours was less than 1.9 mg/l.

7. Methods and Materials

   A) Culture water was prepared by reconstituting deionized water and filtering it through and Amberlite XAD-7 resin column to remove any potential organic contaminants. This water had a total hardness and alkalinity as calcium carbonate ($CaCO_3$) of 175±15 mg/l and 130±10 mg/l, a dissolved oxygen (DO) concentration of greater than 5.3 mg/l, a specific conductance of 400-600 umhos/cm, a pH range of 7.9-8.3 and a temperature of 22±1°C. The toxicity tests were conducted in 250 ml beakers each containing 150 ml of test solution. Fifteen daphnids, <24 hours old, were impartially distributed to each concentration (5 daphnids per replicate). Concentration levels and percentage mortality are listed in table 1.

Table-1. Concentrations tested and corresponding observed percentage mortalities or Daphnia magna exposed to Roundup.

Nominal Concentration	Percentage Mortality	
mg/l	24 hours	48 hours
24.0	40	100
14.0	53	100
8.7	7	93
5.3	0	33
3.1	7	20
1.9	0	0
control	0	0

B) Statistical Analysis: Moving Average Angle Method.

8. Discussion: This study appears to follow the EPA Guideline requirements for an acute aquatic LC_{50} test. The no discernible effect concentration through 48-hours was less than 1.9 mg/l.

Validation: Core

MRID 00070893

DATA EVALUATION RECORD

1. Chemical: Glyphosate, S# 103601

2. Test Material: Roundup (41.36% ai)

3. Study Type: 48-hour LC_{50} - Daphnia magna

4. Study ID: LeBlanc, G.A.; Surprenant, D.C.; Sleight, B.H. III (1980) Acute Toxicity of Roundup to the Water Flea (Daphnia magna): Report #BW-80-4-636; Monsanto Study No. BN-80-079. (Unpublished study, including letter dated February 21, 1980, from R. Oleson to Robert B. Foster, received April 2, 1981, under 524-308; prepared by EG and G, Bionomics, submitted by Monsanto Co., Washington, DC; CDL:244749-B).

5. Reviewed by: Dennis J. McLane
Wildlife Biologist
EEB/HED
Signature: Dennis McLane
Date: 8-28-85

6. Approved by: Raymond W. Matheny
Section Head
EEB/HED
Signature: Raymond W. Matheny
Date: 8-30-85

7. Conclusion:

This study can be used for hazard assessment purposes. Also, it meets the guideline requirements for formulated product testing. Using the toxicity categories of Brooks et al. (1973) the acute LC_{50} of 5.3 (4.4 to 6.3) mg/l would place Roundup, 41.36% ai, into the category of moderately toxic.

8. Recommendation:

N/A

9. Background:

This study was first reviewed by M. Rexrode on July 6, 1981.

10. Discussion of Individual Test:

N/A

11. Materials and Methods:

a. Test animals - were Daphnia magna from EG and G, Bionomics Laboratory stocks; less than 24 hours old.

Test system - Two hundred fifty ml beakers per 150 ml of test solution; static exposure to reconstituted water, at 23 °C; 48-hour duration.

b. Dose - Static bioassay using nominal concentrations; no solvent used.

c. Design - Fifteen Daphnia per level; 6 dose levels plus control (1.9, 3.1, 5.3, 8.7, 14, and 24 mg/l).

d. Statistics (Excerpted from the Study):

Stephan, C. (1978) U.S. EPA, Environmental Research Laboratory, Duluth, MN, Personal communication.

12. Reported Results (Excerpted from the Study):

Table 1. Estimated LC_{50} values and confidence intervals for the water flea (Daphnia magna) exposed to Roundup.

LC_{50} (mg/l)		No-discernible-effect concentration through 48 hours (mg/l)
24 hour[a]	48 hour[b]	
24	5.3 (4.4 - 6.3)[c]	< 1.9

[a]Estimated by probit analysis.
[b]Estimated by the moving average angle method.
[c]95% confidence interval.

13. Study Author's Conclusions/QA Measures (Excerpted from Study):

The 48-hour LC_{50} for the water flea exposed to Roundup, estimated by the moving average angle

method, was 5.3 mg/l. Table 1 summarizes the estimated LC_{50}'s and confidence intervals and states the no-discernible-effect concentration through 48 hours. The no-discernible-effect concentration is the highest concentration tested at which there were no mortalities or observed behavioral and physical abnormalities (i.e., erratic swimming, flared carapace).

14. Reviewer's Discussion and Interpretation of the Study:

a. Test Procedures: The study procedures follow the guidelines if formulated product testing is required.

b. Statistical Analysis: The EEB Lexitron Probit method agrees with the reported value of 5.3 mg/l.

c. Discussion/Results: The study is adequate for hazard assessment and meets the guideline for formulated product, Roundup.

d. Adequacy of Study:

1. Classification: Core for the formulated product.

2. Rationale: The study followed the guideline procedures.

3. Repairability: N/A

15. Completion of One-Liner for Study:

Completed July 12, 1985

16. CBI Appendix:

N/A

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3. Reviewed by: Miachel Rexrode
Fishery Biologist
Ecological Effects Branch, HED

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7. Methods and Materials

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Validation: Core

00070893

MCLANE GLYPHOSATE DAPHNIA 48 HOUR LC50

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB.(PERCENT)
24	15	15	100	3.05176E-03
14	15	15	100	3.05176E-03
8.7	15	14	93.3333	.0488281
5.3	15	5	33.3333	15.0879
3.1	15	3	20	1.75781
1.9	15	0	0	3.05176E-03

THE BINOMIAL TEST SHOWS THAT 3.1 AND 8.7 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 5.99845

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
4	.0707891	5.1664	4.29286	6.18928

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
5	.13801	1	.423686

SLOPE = 5.44716
95 PERCENT CONFIDENCE LIMITS = 3.42356 AND 7.47077

LC50 = 5.24957
95 PERCENT CONFIDENCE LIMITS = 4.33937 AND 6.32356

LC10 = 3.06884
95 PERCENT CONFIDENCE LIMITS = 2.07608 AND 3.80356

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INERT INGREDIENT INFORMATION IS NOT INCLUDED

DATA EVALUATION RECORD

1. CHEMICAL: Glyphosate

2. FORMULATION: This is the "W" formulation which contains [redacted] as a surfactant (MON 2139 NF-80W).

3. CITATION: Forbis, A.D., Boudreau, P. 1980. Acute toxicity of MON 2139 NF-80W (AB-80-365) to Daphnia magna Analytical Bio Chemistry Laboratories, Inc. Submitted by Monsanto Co. CDL Acc #070171, Part C for EPA Reg. No. 524-308, Petition Numbers 9F2163, 9H5204 on 7-1-81

4. REVIEWED BY: Dennis J. McLane
Biologist
EEB/HED

5. DATE REVIEWED: 8-3-81

6. TEST TYPE: Forty-eight hour LC_{50} on Daphnia magna

7. REPORTED RESULTS:

Compound	48-hour LC_{50} (95% C.I.)
MON 2139 NF-80W	72 (62-83) mg/l

8. REVIEWER'S CONCLUSIONS:

This study is scientifically sound and indicates that the compound is slightly toxic to Daphnia magna.

9. MATERIALS AND METHODS

A. Test Procedures

Five concentrations of the test compound, 32, 56, 100, 180, and 320 mg/l, with ten Daphnia per concentration were selected for definitive bioassay.

B. Statistical Analysis

The statistical values were obtained by employing a computerized LC_{50} program developed by Stephan's EPA.

C. Discussion/Results

The following chart of toxic reactions was presented in the raw data:

		24 hrs		48 hrs	
Test Conc.		Dead	Obs	Dead	Obs
Control	a	0	N	0	N
	b	0	N	0	N
32 mg/l	a	0	N	0	N
	b	0	N	0	N
56	a	0	2 N 8 OTB	4	6 OTB
	b	0	2 N 8 OTB	2	8 OTB
100	a	4	6 OTB	9	1 OTB
	b	4	6 OTB	7	3 OTB
180	a	8	2 OTB	10	-
	b	8	2 OTB	10	-
320	a	9	1 OTB	10	-
	b	8	2 OTB	10	-

N = Normal OTB = On the Bottom

10. REVIEWER'S EVALUATION

A. Test Procedure

The test procedure was scientifically sound.

B. Statistical Analysis

The reported statistical values were identical to those produced by the binomial method available to EEB. (see attached computer printout.)

C. Discussion/Results

The testing of a formulation is not covered by the guidelines. However, this study is scientifically sound and is sufficient for use in a hazard assessment.

D. Conclusion

Supplemental

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB.(PERCENT)
320	20	17	85	0.1288414
180	20	16	80	0.5908966
100	20	8	40	25.17223
56	20	0	0	9.536743E-05
32	20	0	0	9.536743E-05

THE BINOMIAL TEST SHOWS THAT 56 AND 180 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 115.1136

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
3	0.08602108	130.2456	106.3672	158.5978

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
6	0.1053849	1	0.128189

SLOPE = 3.982357
95 PERCENT CONFIDENCE LIMITS = 2.689563 AND 5.275151

LC50 = 134.0922
95 PERCENT CONFIDENCE LIMITS = 109.9532 AND 165.1647

LC10 = 64.34237
95 PERCENT CONFIDENCE LIMITS = 42.9416 AND 81.90297
****

? 16
? 6
? 0

CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
320	20	20	100	9.536743E-05
180	30	20	100	9.536743E-05
100	20	16	80	0.5908966
56	20	6	30	5.765915
32	20	0	0	9.536743E-05

THE BINOMIAL TEST SHOWS THAT 32 AND 100 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 70.25257

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
3	0.05105427	71.79532	61.67521	83.20347

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
3	0.141968	1	0.8664117

SLOPE = 6.465971
95 PERCENT CONFIDENCE LIMITS = 4.029681 AND 8.902261

LC50 = 70.92865
95 PERCENT CONFIDENCE LIMITS = 60.52208 AND 83.25993

LC10 = 45.12413
95 PERCENT CONFIDENCE LIMITS = 32.53717 AND 58.96948

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

3 NOV 1983

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

TO: EEB Files

THRU: Raymond Matheny RWM
Head, Section #1
Ecological Effects Branch
Hazard Evaluation Division, TS-769

SUBJECT Previously submitted Aquatic Studies with Glyphosate and the Surfactants "W", "AA", "X-77" as well as 62.4% Glyphosate, isopropylamine (IPA) salt with No surfactant.

The percentage of active ingredient was not reported for the subject studies. Hence, Lyle Gingericil (223-6968) of Monsanto's Washington Office was contacted and the formulation information requested. The percentage of both the IPA salt of glyphosate and surfactant were available.

	IPA salt of glyphosate	Surfactant
1.	40.7% (MON2139)*	"W" - 15% DM
2.	41.2% (MON2139)	"AA" - 15.3%
3.	7.03% (MONO139)**	"X-77" - 0.5%
4.	62.4% (MONO139)	No Surfactant

* MON2139 is the Roundup formulation according to previous EEB reviews

** MONO139 is the isopropylamine salt of glyphosate

Dennis McLane

Dennis J. McLane
Wildilfe Biologist
Hazard Evaluation Division, (TS-769c)