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DATA EVALUATION RECORD § 70-1

CHEMICAL: Glyphosate <u>S#.</u>: 417300 or 103601

TEST MATERIAL: Technical Glyphosate and Roundup® 360 <u>Purity</u>: Not reported

3. CITATION

> <u>Authors:</u> Joseph R. Bidwell and John R. Gorrie

<u>Title:</u> Acute Toxicity of a Herbicide to Selected

Frog Species Final Report Curtin

Ecotoxicology Program, Curtin University

of Technology, Unit 7, R&D Centre,

Technology Park, Bentley 6102

Study Completion Date: June 1995 Laboratory: (see above)

Western Australian Department of Sponsor:

Environmental Protection,

Westralia Square, 141 St. George's

Terrace, Perth, 6000

N/A<u>Laboratory Report ID:</u>

MRID No.: 43839601 <u>DP Barcode</u>: D226164

Depais J. McLane, Wildlife, EEB, EFED REVIEWED BY:

Signature:

Date: 4-17-77

APPROVED BY: Les W. Touart, Head of Section 1, EEB, EFED

Signature:

Date:

STUDY PARAMETERS

Scientific Name of Test Organism: Crinia insignifera,

Litoria moorei

Age or Size of Test Organism: Adult: C. insignifera

> Newly emerged: C.insignifera

Tadpoles: C.insignifera &

L. moorei

Static

Definitive Test Duration: 24, 48, 72, 96 hours

Study Method: Type of Concentrations: Nominal

CONCLUSIONS:

The study is scientifically sound but EPA does not have a guideline requirement for frog testing. Based on Brooks (1973) categories of toxicities, the lowest LC50 of 7.6 mg/L for L.moorei tadpoles, would be "moderately toxic" (.1-10 mg/L).



Results Synopsis

(In 12. Reported Results see tables 3, 6, and 8)

8. ADEQUACY OF THE STUDY

- A. Classification: Supplemental
- B. Rationale: Lack of frog testing requirement.
- C. Repairability: N/A

9. GUIDELINE DEVIATIONS

N/A

10. SUBMISSION PURPOSE:

6(A)2 submission made in connection with this study and Monsanto's response.

11. MATERIALS AND METHODS

11.1 Source of test animals

"Frogs and tadpoles were collected from sites within the Perth metropolitan area and as far south as Mandurah, Western Australia (~80 km from Perth)."

"Bioassays were initiated within two days of receiving the frogs in order to minimis stress associated with being held in the laboratory. Test protocols were reviewed and approved by the Curtin University of Technology Animal Experimentation Ethics Committee."

11.2 Test protocol

"Adult C. insignifera (15 to 23 mm S-V, Snout-vent length) and L.adeaidensis (37 to 43 mm S-V) were tested in chambers which facilitated individual exposure to test solutions (Figure 1). The exposure chambers were constructed from short lengths (15 mm for C. insignifera, 30 mm for L. adelaidensis) of PVC pipe (50 mm diameter) which were covered on one end with 0.5 mm nylon mesh. The opposite side was fitted with the lid of a 55 mm polystyrene petri dish. Bioassays were initiated by placing the chambers into glass beakers which contained the test solutions. The chambers were placed in the beakers with the diameter vertical, allowing the meshed side to remain unobstructed and facilitating passage of test solution and air. Bioassays with C. insignifera were conducted in 250 ml beakers containing 125 ml of test solution.

This provided a 15 mm air space within the exposure chamber. Tests with the larger *L. adelaidensis* were similarly conducted in 250-ml beakers, but with only 100 ml of test solution to allow a 20 mm air space. When undisturbed, the frogs would usually cling to the mesh side of the exposure chamber in the airspace. The volumes of test solution were chosen so that even in this resting position the animals would have some skin contact with the solution. Daily renewal of the bioassay was achieved by pouring off the old test solution and replacing it with new."

"Adult L/ moorei were too large (40 to 70 mm S-V) for the test chambers described above and so were exposed to the herbicides in replicate 5 l glass beakers containing 200 ml of test solution and holding three frogs each (n=6). The test solution was 1.5 cm deep and largely covered the hindquarters of a frog sitting upright. Each beaker covered with a Perspex plate through which a number of air holes were drilled."

"In contrast to *L. moorei*, newly emerged *c. insignifera* ranged 5 to 11 mm S-V and were too small to be effectively exposed in the test chambers used for full size adults. The bioassay with these frogs was conducted by placing individuals in glass test tubes (30 mm diameter x 120 mm length) with a 0.5 mm nylon mesh across one end. A large swab of glass fiber was inserted through the top of the tube, creating a chamber 10 to 15 mm high. Racks holding 10 tubes were then suspended in two replicates 3 L beakers containing enough test solution to immerse the tubes 5 mm deep. A frog sitting upright in a tube was partly covered in test solution without having to continuously swim. Daily renewal of the bioassay was accomplished by transferring the racks of tubes to alternate beakers containing fresh solution."

"Tests with *C. insignifera* and *L. moorei* tadpoles were conducted in 250 ml glass beakers containing 100 ml of test solution. Each beaker held five tadpoles with two or three replicates per concentration."

"Daily renewals of the bioassay were achieved by first draining the old test solution over a nylon mesh (in the event a tadpole was accidentally poured out during the change over) and replaced with new."

"All bioassay were conducted at $20\pm2\,^{\circ}\text{C}$ under ambient laboratory light."

11.3 Test substance

"Technical grade glyphosate (N-(Phosphonomethyl) Glycine, 96%) was obtained from a local supplier on 23 June 1994. The formulated herbicide "Roundup 360" was purchased from a local retail outlet."

A 1 mg/ml stock solution of glyphosate was prepared by adding 1 g of powdered chemical to 1 L of distilled water. Levels of Roundup 360 were calculated and reported as glyphosate active (isopropylamine salt). Test solutions were prepared by serial dilution of the highest concentration tested. Test concentrations for initial screening studies were based on suggested application levels of the herbicide."

"Purity of the technical grade glyphosate and analyses of selected test solutions were conducted by the Chemistry Centre (WA) using high performance liquid chromatography (HPLC)."

11.4 Diluent

"Aged (three days), aerated laboratory tap water was used ass the diluent in the screening tests with *C. insignifera*, *L. adelaidensis*, and *L. moorei*. All other bioassay used filtered (30 mm) water collected from a lake on the Curtin University campus."

11.5 Statistical analyses

"Mortality data were used to generate LC50 values by the Spearman-Karber method (Hamilton et al. 1978). Reported values are based on target (nominal) rather than measured concentrations of glyphosate."

12. REPORTED RESULTS

A. General Results

Table 2. Toxicity of technical grade glyphosate and Roundup 360 (expressed as glyphosate active) to adult *C. insignifera* in a screening study conducted between 11 and 15 August 1994.

Concentration (ppm)	Number	Cum	ılative I	Number D	ead
	of Frog		Hour of	Study	
Nominal		24	48	72	96
Control	5	.0	0	20	20
Roundup 360 11.1	5	0	0	0	0
Glyphosate 22.3	5	0	0	0 -	0
Roundup 360 22.3	5	0	0	0	0

Concentration (ppm)	Number	Cumulative Number Dead				
	of Frog	*	Hour of	Study		
Nomina1		24	48	72	96	
Glyphosate 45	5	0	0	0	0	
Roundup 360 45	5	20	20	40	60	
Glyphosate 67.6	5	20	40	40	40	
Roundup 360 67.6	5	100				
Glyphosate 90	5	40	40	60	60	
Roundup 360 90	5	80	100		2.3	
Glyphosate 135	5	100			· · · · · · ·	

Table 3. Preliminary LC50 values (95% confidence interval) for adult *C. insignifera* in screening bioassays with glyphosate or Roundup 360 (expressed as glyphosate active).

Time		LC50
	Glyphosate	Roundup 360
24 h 48 h 72 h	89.6 (73.6-108.9) 83.6 (67.4-103.6) 72.0 (62.9-96.6)	52.6 (39.3-70.5) 49.4 (40.5-60.2) 44.2 (34.7-56.3)
96 h	78.0 (62.9-96.6)	39.7 (31.1-50.5)

Table 4. Toxicity of technical grade glyphosate and Roundup 360 (expressed as glyphosate active) in a screening test conducted between 21 and 25 September 1994.

Table 5. Toxicity of technical grade glyphosate and Roundup 360 (expressed as glyphosate active) in a screening test with *C. insignifera* tadpoles.

Toxica	ant		Cumulative	% Mortality	Y
(mg/L)	# Exposed	24h	48h	72 h	96 h
0	10	0	0	0	0
Glyphosate 22.5	10	0	,0	0	0
Roundup 360 22.5	10	20	100		
Glyphosate 45	10	0	0	0 -	.0
Roundup 360 45	10	100			
Glyphosate 67.5	10	0	0	0	0
Roundup 360 67.5	10	100			
Glyphosate 90	10	0	0	0	0
Roundup 360 90	10	100			
Glyphosate 180	10	0	0	0	0 -
Roundup 360 180	10	100			

Table 6. Toxicity of Roundup 360 (expressed as glyphosate active) to newly emerged *C.insignifera*.

Roundup 360		Cumula	tive % Mort	ality
(mg/L)	# Exposed	24h	48h	/72 h
0	20	5	10	20
11.5	20	0	5	15
22.5	20 '	0	5	10
45	20	3.0	40	65
90	20	35	85	100
180	20	90	100	
LC5 (95% conf interv	idence	88.7 (68.14.6)	51.8 (42.1- 63.8)	

^{* -} no LC50 calculated due to control mortality.

Toxicant		Cumulative % Mortality				
(mg/L)	# Exposed	24h	48h	72 h	96 h	
0	15	0	0	`0	Ó	
Roundup 360 1.12	15	0	0	0	0	
Roundup 360 2.25	15	0	13	0	0	
Roundup 360 4.5	15	0	0.	0	20	
Roundup 360 9.0	15	0		40	53	
Glyphosate 11.25	15	0	0	0	0	

Roundup 360 18.0	15	100			
Glyphosate 22.5	15	0	0	7	13
Roundup 360 36	15	100	7,		
Glyphosate 45	15	0		0	0
Glyphosate 90	15	0		13	27
Glyphosate 180	15	100			

Table 8. LC50 values (95% confidence intervals) for *L. moorei* tadpoles exposed to technical grade glyphosate or Roundup 360 (expressed as glyphosate active).

Time	LC50 (mg/L)
	Glyphosate Roundup 360
24 h 48 h 72 h 96 h	127.0 (90.0-180.0) 12.7 (9.0-18.0) 121.5 (111.2-132.9) 11.6 (10.3-13.1) 116.0 (102.2-131.8) 10.6 (9.0 - 12.4) 110.8 (95.2-128.4) 7.66 (6.1 - 9.6)

[&]quot;.... The presence of a surfactant in the formulation that was tested may then explain why tadpoles which respire with gills, were significantly more sensitive to Roundup 360 than adult frogs."

13. VERIFICATION OF STATISTICAL RESULTS

The following tables was excerpted from the study (B&G95) and the values recalculated using toxanal (EPA).

Table 3. Preliminary LC50 values (95% confidence interval) for adult *C. insignifera* in screening bioassays with glyphosate or Roundup 360 (expressed as glyphosate active).

Time	I	C50
	Glyphosate	Roundup 360
24 h 48 h 72 h 96 h	EPA 83.1 (61.7-116.1) B&G95 72.0 (62.9-96.6) EPA 78 (57.5-105.8)	

Reported values

Table 6. Toxicity of Roundup 360 (expressed as glyphosate active) to newly emerged *C.insignifera*.

ī					
	Roundup 360		Cumulat	ive % Mortality	
	(mg/L)	# Exposed	,24h	48h	72 h
	0	20	5	10	20
-	11.5	20	0	5	15
	22.5	20	0	5	10
	45	20	30	40	65
	90	20	35	85	100
	180	20	9.0	100	-
	LC5((95% conf interv	idence	B&G95 88.7 (68.6- 114.6) EPA 87.8 (69-115.7)	B&G95 51.8 (42.1-63.8) EPA 51.6 (40.8-65.9)	*

^{* -} no LC50 calculated due to control mortality.

Table 8. LC50 values (95% confidence intervals) for L. moorei tadpoles exposed to technical grade glyphosate or Roundup 360 (expressed as glyphosate active).

Time	LC5	0 (mg/L)
	Glyphosate	Roundup 360

² EPA calculated values

24 h	B&G95 127.0 (90.0-180.0) 12.7 (9.0-18.0)
	EPA 127.3 (90-180) 12.7 (9-18)
48 h	B&G95 121.5 (111.2-132.9) 11.6 (10.3-13.1)
	EPA 92.9 (45-180) 11.6 (9-18)
72 h	B&G95 116.0 (102.2-131.8) 10.6 (9.0 - 12.4)
	EPA 116.0 (90-180) 9.8 (4.5-18)
96 h	B&G95 110.8 (95.2-128.4) 7.66 (6.1 - 9.6)
	EPA 107.5 (45-180) 7.6 (6.0-9.6)

14. REVIEWER'S COMMENTS:

The statistical methods were acceptable.

The control mortality in the adult *C. insignifera* test for the 72 hour and 96 hour was 20%. The two day acclimation period was probably not adequate to determine the health of the *C.insignifera* population. Newly emerged *C.insignifera* also had the same problem with control mortality 5% at 24 hours, 10% at 48 hours, and 20% at 72 hours. A longer acclimation period may have provided more about the health and husbandry of *C.insignifera*.

Roundup 360 is 41% glyphosate the same a Roundup in this country. As cited by the authors Folmar (et al. 1979) has shown that the surfactant increased the toxicity of the product. The lowest LC50 derived for glyphosate in this study was 72 mg/L (62.9-96.6) mg/L versus 7.6 mg/L for Roundup 360.

The study would indicate that tadpoles are the most sensitive to Roundup 360 with an 96 hour *L.moorei* LC50 of 7.6 mg/L versus 96 hour LC50 *C.insignifera* of 39.7 mg/L for adults and 48 hour LC50 of 51.5 mg/L for newly emerged (*L.moorei*) (96 hours for newly emerged was not available).

The screening study with Roundup 360 shows that *C.insignifera* tadpoles may also be as sensitive as *L.moorei*. All of the *C.insignifera* tadpoles died after 48 hours in a concentration of 22.5 mg/L.

MCLANE ROUNDUP 360 TADPOLES 24 HOUR LC50

*****	*****	******	*****	*******
CONC.	NUMBER	NUMBER	PERCENT	BINOMIAL
	EXPOSED	DEAD	DEAD	PROB. (PERCENT)
36	15	15	100	3.051758E-03
18	15	15	100	3.051758E-03
9	15	0	0	3.051758E-03
4.5	15	- 0		3.051758E-03
2.25	15	0	0	3.051758E-03
1.12	15	0	0	3-051758E-03

THE BINOMIAL TEST SHOWS THAT 9 AND 18 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 12.72792

WHEN THERE ARE LESS THAN TWO CONCENTRATIONS AT WHICH THE PERCENT DEAD IS BETWEEN 0 AND 100, NEITHER THE MOVING AVERAGE NOR THE PROBIT METHOD CAN GIVE ANY STATISTICALLY SOUND RESULTS.

MCLANE ROUNDUP 360 TADPOLES 48 HOUR LC50

3.051758E-03 18 15 15 100 15 2 13.33333 .3692627 4.5 15 0 0 . 3.051758E-03 2.25 15 0. 3.051758E-03 1.12 15 3.051758E-03

THE BINOMIAL TEST SHOWS THAT 9 AND 18 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 11.6006

WHEN THERE ARE LESS THAN TWO CONCENTRATIONS AT WHICH THE PERCENT DEAD IS BETWEEN 0 AND 100, NEITHER THE MOVING AVERAGE NOR THE PROBIT METHOD CAN GIVE ANY STATISTICALLY SOUND RESULTS.

MCLANE	ROUNDUP	360 I	ADPOLES	72 HOU	R LC50			
*****	******	****	*****	****	*****	****	*****	*****
CONC.	NUMBER		NUMBER		PERCENT	BINOMIA	L.	
	EXPOSE)	DEAD		DEAD	PROB. (I	ERCENT)	

36 15 15 100 3.051758E-03 18 15 15 100 3.051758E-03

· 9 h	15	6	40	30.36194
4.5	15	0	0	3.051758E-03
2.25	15	0	0	3.051758E-03
1.12	15	.0	0	3.051758E-03

THE BINOMIAL TEST SHOWS THAT 4.5 AND 18 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 9.817339

WHEN THERE ARE LESS THAN TWO CONCENTRATIONS AT WHICH THE PERCENT DEAD IS BETWEEN 0 AND 100, NEITHER THE MOVING AVERAGE NOR THE PROBIT METHOD CAN GIVE ANY STATISTICALLY SOUND RESULTS.

MCLANE ROUNDUP 360 TADPOLES 96 HOUR LC50

********************************** PERCENT NUMBER NUMBER BINOMIAL DEAD EXPOSED DEAD PROB. (PERCENT) 15 15 100 3.051758E-03 100 18 15 15 3.051758E-03 15 8 53.33334 50 9 3 (4.5 15 20 1.757813 2.25 15 0 0 3.051758E-03 1.12 15 3.051758E-03

THE BINOMIAL TEST SHOWS THAT 4.5 AND 18 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 8.428467

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD
SPAN G LC50 95 PERCENT CONFIDENCE LIMITS
5 7.146491E-02 7.326024 5.533304
9.912052

RESULTS CALCULATED USING THE PROBIT METHOD ITERATIONS G H
GOODNESS OF FIT PROBABILITY
6 .168964 1
.755761

SLOPE = 4.833743

95 PERCENT CONFIDENCE LIMITS = 2.846822 AND 6.820664

LC50 = 7.55571 95 PERCENT CONFIDENCE LIMITS = 5.95973 AND 9.580359

LC10 = 4.126093

MCLANE GLYPHOSATE TADPOLES 24 HOUR LC50

*****	*****	*****	******	*******	
CONC.	NUMBER	NUMBER	PERCENT	BINOMIAL	
	EXPOSED	DEAD	DEAD	PROB. (PERCENT)	
180	15	15	100	3.051758E-03	
90	15	0	0	3.051758E-03	
45	15	, 0		3.051758E-03	
22.5	15		0	3.051758E-03	
11.25	15		0	3.051758E-03	

THE BINOMIAL TEST SHOWS THAT 90 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 127.2792

WHEN THERE ARE LESS THAN TWO CONCENTRATIONS AT WHICH THE PERCENT DEAD IS BETWEEN 0 AND 100, NEITHER THE MOVING AVERAGE NOR THE PROBIT METHOD CAN GIVE ANY STATISTICALLY SOUND RESULTS.

MCLANE GLYPHOSATE TADPOLES 48 HOUR LC50

CONC.	NUMBER	NUMBER	PERCENT	BINOMIAL
	EXPOSED	DEAD	DEAD	PROB. (PERCENT)
180	15	15	100	3.051758E-03
90	15	7	46.66667	10 (50)
45	15 0 / / / / / /		0	3.051758E-03
22.5	15	0	0	3.051758E-03
11.25	15	0 .	0	3.051758E-03

THE BINOMIAL TEST SHOWS THAT 45 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 92.87608

WHEN THERE ARE LESS THAN TWO CONCENTRATIONS AT WHICH THE PERCENT DEAD IS BETWEEN 0 AND 100, NEITHER THE MOVING AVERAGE NOR THE PROBIT METHOD CAN GIVE ANY STATISTICALLY SOUND RESULTS.

MCLANE GLYPHOSATE TADPOLES 72 HOUR LC50

CONC. NUMBER EXPOSED

NUMBER DEAD PERCENT DEAD

BINOMIAL PROB. (PERCENT)

180 15	15	1	.00	3.051758E-03
90 15	2	. 1	3.33333	.3692627
45 15	0	0		3.051758E-03
22.5 15		6	.666667	4.882813E-02
11.25 15	0	0		3.051758E-03

THE BINOMIAL TEST SHOWS THAT 90 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 116.006

THE MOVING AVERAGE METHOD CANNOT BE USED WITH THIS DATA SET BECAUSE NO SPAN WHICH PRODUCES MOVING AVERAGE ANGLES THAT BRACKET 45 DEGREES ALSO USES TWO PERCENT DEAD BETWEEN 0 AND 100 PERCENT.

RESULTS CALCULATED USING THE PROBIT METHOD
ITERATIONS G
GOODNESS OF FIT PROBABILITY
8 7.593905 13.79384

A PROBABILITY OF 0 MEANS THAT IT IS LESS THAN 0.001.

SINCE THE PROBABILITY IS LESS THAN 0.05, RESULTS CALCULATED USING THE PROBIT METHOD PROBABLY SHOULD NOT BE USED.

SLOPE = 4.285732 95 PERCENT CONFIDENCE LIMITS =-7.524477 AND 16.09594

LC50 = 106.867 95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

MCLANE GLYPHOSATE TADPOLES 96 HOUR LC50

CONC.	NUMBER	NUMBER	PERCENT	BINOMIAL
	EXPOSED	DEAD	DEAD	PROB. (PERCENT)
180	15	15	100	3.051758E-03
90	15	4	26.66667	5.923462
45	15		0	3.051758E-03
22.5	15	2	13.33333	.3692627
11.25	15	0	0	3.051758E-03

THE BINOMIAL TEST SHOWS THAT 45 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 107.4888

THE MOVING AVERAGE METHOD CANNOT BE USED WITH THIS DATA SET BECAUSE NO SPAN WHICH PRODUCES MOVING AVERAGE ANGLES THAT BRACKET 45 DEGREES ALSO USES TWO PERCENT DEAD BETWEEN 0 AND 100 PERCENT.

RESULTS CALCULATED USING THE PROBIT METHOD ITERATIONS

GOODNESS OF FIT PROBABILITY

3.053788

6.425535

0

A PROBABILITY OF 0 MEANS THAT IT IS LESS THAN 0.001.

SINCE THE PROBABILITY IS LESS THAN 0.05, RESULTS CALCULATED USING THE PROBIT METHOD PROBABLY SHOULD NOT BE USED.

SLOPE 3.368341

95 PERCENT CONFIDENCE LIMITS =-2.517865 AND

9.254546

LC50 = 94.03784

95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

LC10 =39.46956

95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

NOTE: THERE WAS CONTROL MORTALITY, BUT AT LEAST ONE OF THE LOWER CONCENTRATIONS HAD ZERO MORTALITY. THEREFORE, ABBOTT'S CORRECTION IS NOT APPLICABLE.

MCLANE rOOUNDUP 360 NEWLY EMERGED FROGS 24 HOUR LC50

NUMBER CONC. NUMBER PERCENT BINOMIAL DEAD PROB. (PERCENT) EXPOSED DEAD 180 20 18 9.0 2.012253E-02 90 20 7 35 13.1588 20 / 45 6 30 5.765915 22.5 20 0 0 9.536742E-05 11.25 20 0 9.536742E-05

THE BINOMIAL TEST SHOWS THAT 22.5 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 107.0428

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN G LC50

95 PERCENT CONFIDENCE LIMITS

. 3 7.602793E-02 84.87036 68.47653

110.2925

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS

GOODNESS OF FIT PROBABILITY

.1275428 1

.1351399

SLOPE 3.294123

95 PERCENT CONFIDENCE LIMITS = 2.117689 AND 4.470558

LC50 = 87.77271

95 PERCENT CONFIDENCE LIMITS = 69.04436 AND 115.7058

LC10 = 36.12639

95 PERCENT CONFIDENCE LIMITS = 21.61119 AND 48.30736

NOTE: BECAUSE THERE WAS CONTROL MORTALITY, AND NONE OF THE LOWER CONCENTRATIONS PRODUCED ZERO MORTALITY, THE DATA HAS BEEN SUBJECTED TO ABBOTT'S CORRECTION.

MCLANE rOOUNDUP 360 NEWLY EMERGED FROGS 48 HOUR LC50 ******************************

CONC. NUMBER NUMBER PERCENT BINOMIAL EXPOSED DEAD DEAD PROB. (PERCENT) 180 18 18 100 3.814697E-04 9.0 18 15 83.3333 .3768921 45 18 6 33.3333 11.89423 20 22.5 5 2.002716E-03 11.25 20 5 2.002716E-03

THE BINOMIAL TEST SHOWS THAT 22.5 AND 90 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 56.15447

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN G LC50 95 PERCENT CONFIDENCE LIMITS

7.029575E-02 52.40946 41.06064

68.63534

RESULTS CALCULATED USING THE PROBIT METHOD ITERATIONS

GOODNESS OF FIT PROBABILITY

.1092167

.1195951

SLOPE 3.634041

95 PERCENT CONFIDENCE LIMITS = 2.433065 AND 4.835017

LC50 = 51.61713

95 PERCENT CONFIDENCE LIMÍTS = 40.78895 AND 65.87588

LC10 =23.08451

95 PERCENT CONFIDENCE LIMITS = 14.46924 AND 30.49163

NOTE: BECAUSE THERE WAS CONTROL MORTALITY, AND NONE OF THE LOWER CONCENTRATIONS PRODUCED ZERO MORTALITY, THE DATA HAS BEEN SUBJECTED TO ABBOTT'S CORRECTION.

MCLANE rOOUNDUP 360 NEWLY EMERGED FROGS 72 HOUR LC50

****************************** NUMBER NUMBER PERCENT BINOMIAL EXPOSED DEAD DEAD PROB. (PERCENT) 180 16 ,16 100 1.525879E-03 16 16 90 100 1.525879E-03 . 9 45 16 56.25 40.18097 20 2 10 22.5 2.012253E-02 11.25 20 15 .1288414

THE BINOMIAL TEST SHOWS THAT 22.5 AND 90 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 41.41958

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN G LC50 95 PERCENT CONFIDENCE LIMITS

4 .1027428 33.85614 23.8361 45.42646

RESULTS CALCULATED USING THE PROBIT METHOD ITERATIONS G H
GOODNESS OF FIT PROBABILITY

.0278303 .037544

SINCE THE PROBABILITY IS LESS THAN 0.05, RESULTS CALCULATED USING THE PROBIT METHOD PROBABLY SHOULD NOT BE USED.

SLOPE = 3.360031

95 PERCENT CONFIDENCE LIMITS = .1141276 AND 6.605933

LC50 = 35.00115

95 PERCENT CONFIDENCE LIMITS = 3.848137 AND 837.4293

LC10 = 14.65917

95 PERCENT CONFIDENCE LIMITS = 5.241533E-10 AND 29.41597

MCLANE ROUNDUP 360 FROG 24 HOUR LC50

CONC.	NUMBER NUMBER	PERCENT	BINOMIAL
	EXPOSED DEAD	DEAD	PROB. (PERCENT)
90	4.	80 (1)	18.75
67.6		100	3.125
45	- 5 1 1 1 2 4 4 1 1 1 1 1 1 1 1 1 1 1 1 1 1	20	18.75
22.3	6 - 5 - 3 - 3 - 4 - 4 - 4 - 0 - 3 - 4 - 4 - 4 - 4		3.125
11.1	. 5	0	3.125

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 51.20384

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN LC50 G 95 PERCENT CONFIDENCE LIMITS 20.07521 86.09863 .5976201 44.42308

RESULTS CALCULATED USING THE PROBIT METHOD ITERATIONS G

GOODNESS OF FIT PROBABILITY

.6470405

.3455239

SLOPE 6.741876

95 PERCENT CONFIDENCE LIMITS = 1.31879 AND 12.16496

LC50 = 54.23584

95 PERCENT CONFIDENCE LIMITS = 29.41832 AND 73.9661

LC10 = 35.1489

11.1

95 PERCENT CONFIDENCE LIMITS = 3.914442 AND 47.59696

MCLANE ROUNDUP 360 FROGS 48 HOUR LC50

*********************** PERCENT CONC. NUMBER NUMBER BINOMIAL EXPOSED DEAD DEAD PROB. (PERCENT) - 5 9.0 5 100 3.125 67.6 5 5 100 3.125 45 1 20 18.75 22.3

0

3.125

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 51.20384

0

WHEN THERE ARE LESS THAN TWO CONCENTRATIONS AT WHICH THE PERCENT DEAD IS BETWEEN 0 AND 100, NEITHER THE MOVING AVERAGE NOR THE PROBIT METHOD CAN GIVE ANY STATISTICALLY SOUND RESULTS.

NOTE: THERE WAS CONTROL MORTALITY, BUT AT LEAST ONE OF THE LOWER CONCENTRATIONS HAD ZERO MORTALITY. THEREFORE, ABBOTT'S CORRECTION IS NOT APPLICABLE.

MCLANE ROUNDUP 360 FROGS 72 HOUR LC50

CONC.	NUMBER	NUMBER	PERCENT	BINOMIAL
	EXPOSED	DEAD	DEAD	PROB. (PERCENT)
9.0	5	5	100	3.125
67.6	5	5	100	3.125
45	5	2	40	50
22.3,	5	0	0	3.125
11.1	1.5	0	0	3.125

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 47.41979

WHEN THERE ARE LESS THAN TWO CONCENTRATIONS AT WHICH THE PERCENT DEAD IS BETWEEN 0 AND 100, NEITHER THE MOVING AVERAGE NOR THE PROBIT METHOD CAN GIVE ANY STATISTICALLY SOUND RESULTS.

NOTE: THERE WAS CONTROL MORTALITY, BUT AT LEAST ONE OF THE LOWER CONCENTRATIONS HAD ZERO MORTALITY.
THEREFORE, ABBOTT'S CORRECTION IS NOT APPLICABLE.

MCLANE ROUNDUP 360 FROGS 96 HOUR LC50

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CONC.	NUMBER	NUMBER	PERCENT	BINOMIAL
	EXPOSED	DEAD	DEAD	PROB. (PERCENT)
90	5	6 5	100	3.125
67.6	5	5 5 6 6	100	3.125
45	5	3	60.00001	50
22.3	. 5	0	0	3.125
11.1	5	0	0	3.125

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 41.11195

WHEN THERE ARE LESS THAN TWO CONCENTRATIONS AT WHICH THE PERCENT DEAD IS BETWEEN 0 AND 100, NEITHER THE MOVING AVERAGE NOR THE PROBIT METHOD CAN GIVE ANY STATISTICALLY SOUND RESULTS.

NOTE: THERE WAS CONTROL MORTALITY, BUT AT LEAST ONE OF THE LOWER CONCENTRATIONS HAD ZERO MORTALITY. THEREFORE, ABBOTT'S CORRECTION IS NOT APPLICABLE.

MCLANE GLYPHOSATE FROGS 24 HOUR LC50

************************ PERCENT BINOMIAL NUMBER NUMBER EXPOSED DEAD DEAD PROB. (PERCENT) 135 5 5 100 3.125 2 90 40 50 67.6 1 1 20 18.75 0 45 Ω. 3.125 22.3 5 0 3.125

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 94.82151

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN G LC50 95 PERCENT CONFIDENCE LIMITS

3 .2872567 87.93612 70.43003 120.9225

RESULTS CALCULATED USING THE PROBIT METHOD ITERATIONS G H
GOODNESS OF FIT PROBABILITY
23 .6129955 1

SLOPE = 9.744269

.8559464

95 PERCENT CONFIDENCE LIMITS = 2.115089 AND 17.37345

LC50 = 88.34733 95 PERCENT CONFIDENCE LIMITS = 67.82111 AND 124.6519

NOTE: THERE WAS CONTROL MORTALITY, BUT AT LEAST ONE OF THE LOWER CONCENTRATIONS HAD ZERO MORTALITY.
THEREFORE, ABBOTT'S CORRECTION IS NOT APPLICABLE.

MCLANE GLYPHOSATE FROGS 48 HOUR LC50

CONC. NUMBER NUMBER PERCENT BINOMIAL EXPOSED DEAD DEAD PROB. (PERCENT)

135 5	5	100	3.125
90 5	2	40	50
67.6 5	2	40	50
45 5	0	0	3.125
22.3 5	0	0	3.125

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 94.82151

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN G LC50 95 PERCENT CONFIDENCE LIMITS

3 .2717878 82.50613 65.71425 108.014

RESULTS CALCULATED USING THE PROBIT METHOD ITERATIONS G H
GOODNESS OF FIT PROBABILITY
6 .5468783 1
.5871211

SLOPE = 7.790444

95 PERCENT CONFIDENCE LIMITS = 2.029316 AND 13.55157

LC50 = 83.14075

95 PERCENT CONFIDENCE LIMITS = 61.71223 AND 116.1075

LC10 = 57.12072

95 PERCENT CONFIDENCE LIMITS = 18.96722 AND 72.05848

NOTE: THERE WAS CONTROL MORTALITY, BUT AT LEAST ONE OF THE LOWER CONCENTRATIONS HAD ZERO MORTALITY. THEREFORE, ABBOTT'S CORRECTION IS NOT APPLICABLE.

MCLANE GLYPHOSATE FROG 72 HOUR LC50

S F		EXPOSED	DEAD	DEAD		PROB. (PERC
	135	5	5	100		3.125
	90	5	. 3	60.00	0001	50
	67.6	5	2	40		50
	45	5	0	0		3.125
	22.3	5	0	0		3.125

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 77.99998

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

 SPAN
 G
 LC50
 95 PERCENT CONFIDENCE LIMITS

 3
 .2863572
 77.98539
 60.46964
 100.5498

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS

GOODNESS OF FIT PROBABILITY

.5813513

.8645456

SLOPE =

8.745548

95 PERCENT CONFIDENCE LIMITS = 2.077383 AND 15.41371

LC50 = 77.99186

95 PERCENT CONFIDENCE LIMITS = 57.4704 AND 105.7446

LC10 = 55.8255

95 PERCENT CONFIDENCE LIMITS = 17.74106 AND 69.33968