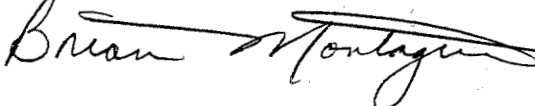
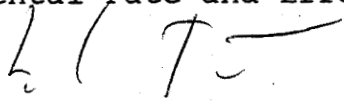


US EPA ARCHIVE DOCUMENT

657201

EEB FILE  
COPYECOLOGICAL EFFECTS BRANCH  
DATA EVALUATION REPORT

1. **Chemical:** Phorate
2. **Test Material:**  $^{14}\text{C}$ -AC35024 Radio purity - 100%
3. **Study Type:** 21-day Chronic Lifecycle Study with *Daphnia magna* under Static Renewal Conditions
4. **Study Identification:**
  - Study Director:** Yurk, J. J.
  - Study Laboratory:** Environmental Science and Engineering, Inc., Gainesville, Florida
  - Study Dates:** July 17 - August 8, 1991
  - Study Identification:** Lab Study No. 3913016-0200-3140
  - Study Sponsor:** American Cyanamid Company
  - EPA Identification:** MRID 422271-02
5. **Reviewed by:** Brian Montague, Fisheries Biologist  
Ecological Effects Branch  
Environmental Fate and Effects Division(H7507C)  
 8/30/93
6. **Approved by:** Les Touart, PhD, Section Supervisor  
Ecological Effects Branch  
Environmental Fate and Effects Division  

7. **Conclusions:** The study has fulfilled data requirements for chronic toxicity testing of phorate technical on a freshwater invertebrate. The results indicate the very high toxicity of this chemical to survival and reproduction of *Daphnia magna* after a 21 day exposure period. Numbers of offspring per female and growth of parental daphnids were both effected at concentrations  $\geq 0.84$  ug/L of phorate. The survival of adult daphnids was effected at concentrations above 0.21 ug/L. The MATC range for chronic effects is  $>0.21 < 0.41$  ug/L based on reduced survival of parental daphnids.
8. **Recommendations:** N/A



9. **Submission Purpose:** Submitted to satisfy freshwater invertebrate full life-cycle testing requirements for Phorate insecticide.
10. **Study Design and Protocol:** American Cynamid's protocol - Study number 941-91-101 "Chronic Toxicity of <sup>14</sup>C AC 35,024 to *Daphnia magna* Under Static - renewal Conditions" used laboratory practices accepted by ASTM in its acute testing guidelines for fish and invertebrates.

**Dilution Water and Test Solutions Preparations:** Dilution water was obtained from an on-site well, passed through a mixed bed deionization resin and Milli Q filter system and reconstituted with 192 mg/L sodium-bicarbonate, 120 mg/L calcium sulfate, 120 mg/L magnesium sulfate and 8 mg/L potassium chloride. Due to low solubility of phorate, stock solutions were prepared in acetone (<0.1 ml/L) and diluted to a concentration of 22.27 mg/L. Based on earlier test results concentrations were prepared at 0.10, 0.25, 0.50, 1.00, and 2.00 ug ai/L from this stock solution.

**Test Organisms:** Test daphnids were obtained from ESE stock cultures maintained in house for five years. Twenty-four hours prior to test initiation gravid females were isolated in clean dilution water. Test organisms were collected from this culture and were <24 hour old neonates. During acclimation cultures were fed *Selenastrum capricornutum*, yeast and trout chow; maintained at 18.7 to 22.4°C, and D.O. levels of 7.8 - 9.0 mg/L.

**Test Materials and Design:** Four replicate 500 ml glass test beakers were used for each control and treatment level. Beakers were filled to a 7 cm depth (400 ml) with dilution water or test solution and housed in a temperature control water bath. Ten daphnids were indiscriminately distributed to each beaker in plastic weigh boats. Beakers were covered with nitex screen. Loading was approximately one daphnid/40 ml of test solution. Up to 24 hours prior to study termination daphnids were fed 0.5 to 3.0 ml of algal mix and 0.25 to 0.75 yeast mixture/day. A 16 day/8 dark photoperiod with 50-minute transition was employed. Daily observations of daphnids were made and mortality, stress indications and numbers of offspring noted. Lengths of 1st generation daphnia were measured at termination and then daphnids oven dried to determine dry weight to nearest 0.1 mg per replicate group. This weight was used to derive average wt./daphnid.

New test solutions were prepared daily. Daphnids were transferred by eye dropper to new test beakers containing the newly-prepared test solutions. Weekly measurement of temperature, D.O., and pH was performed in each replicate of the control or exposure groups. Constant water bath

temperature monitoring was employed. Light intensity, conductivity, alkalinity, and hardness were measured on Day 1 and Day 21 in one control replicate. Pooled samples from each treatment level and control were collected prior to renewal and 0 and 1 day after renewal for concentration analysis by liquid scintillation counting procedures. On days 0, 7, 14, and 21 samples were also removed for analysis by HPLC methods (low, medium, high concentrations only).

11. **Reported Test Results:** Weekly water quality parameters remained within acceptable parameters during the study period. D.O. ranged from 7.2 to 9.1 mg/L, pH from 7.8 to 8.3, and water temperature from 20.0 to 20.9°C. No residues of phorate were detected in control or dilution control test vessels. Measured concentration ranges within each nominal test concentration level were variable, but within acceptable ranges in most instances. Mean average concentrations for the 21 samples (ranges in parenthesis) are as follows: 0.090 (0.062 - 0.124), 0.206 (0.144 - 0.245), 0.405 (0.307 - 0.524), 0.839 (0.659 - 1.057), and 1.805 (1.23 - 2.47) ug ai/L. Detection limit for this method was 0.008 ug/L phorate. HPLC methods yielded average mean values of 0.085, 0.345, and 1.545 ug/L for low, medium and high concentration levels (based on four samples). 21-Day mean percent survival of adult daphnia was 82.5, 80.0, 82.5, 90, 67.5, 45.0 and 2.5 percent for controls, dilution controls, 0.9, 0.21, 0.41, 0.84, and 1.81 ug/L measured concentrations, respectively. The laboratory calculated mean young/adult/reproduction day to be 3.2, 3.5, 3.0, 3.3, 3.2, 3.3, and 1.2 for controls, solvent controls 0.9, 0.21, 0.41, 0.84, and 1.81 ug/L test groups, respectively. Mean dry weight ranged from 0.30 mg for 1.81 ug/L group to 0.49 mg for the 0.21 ug/L group. Controls were 0.41 and 0.4 mg. Mean length ranged from 3.0 mm for the 1.81 ug/L concentration to 3.9 mm for solvent controls.
12. **Study Author's Conclusions:** "The Maximum Acceptable Toxicant Concentration (MATC) of AC 35,024 during the complete life cycle (21 days) of Daphnia magna was 0.59 ug of AC 35,024 equivalents/L based on survival and dry weights. The "no observed effect concentration" (NOEC) of the test material over the 21-day exposure was 0.41 ug of AC 35024/L also based on survival and dry weights."
13. **Reviewer's Discussion:** The study deviated slightly from ASTM accepted protocol in that 10 organisms were housed together in each test vessel, but not separated into individual chambers. Thus, it is not possible to determine accurate individual daphnid reproductive and weight information. It also is not possible to determine if control daphnids were producing minimum numbers of offspring (40 per individual). Estimates of average numbers of offspring produced per daphnid during reproductive days appears to indicate that numbers were above this level.

The mortality of parental daphnids at the 0.41 ug/L concentration level was 32.5% over the 21 day period whereas the combined control mortality was 18.8%. This factor would indicate the NOEL to be below this concentration. The statistical analysis of the mortality values placed an estimated LC10 at 0.12 ug/L using the probit method. Therefore the Agency has determined the NOEL for survival to be 0.21 ug/L, not 0.41 ug/L as stated by the study director.

**Adequacy of Study:**

**Category:** Core

**Rationale:** The study results have demonstrated a defensible MATC level for reproductive/survival effects.

**Repairability:** N/A

**Adequacy of Study**

**Category:**

**Rationale:**

**Repairable:**

Solvent and Dilution  
 Controls inputed  
 80 organisms  
 15 mortalities

18.75%

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

B. Montague Phorate Chronic 21 Day Daphnid

\*\*\*\*\*

CONC.	NUMBER EXPOSED	Actual	NUMBER DEAD	Actual	PERCENT DEAD	% Actual Dead	BINOMIAL PROB. (PERCENT)
1.81	32.5	40	24.5	39	75.3846	97.5%	0
.84	32.5	40	7.5	22	23.0769	55%	0
.41	40	40	6	13	15	32.5%	0
.21	40	40	4	4	10	10%	0
9.000001E-02		40	40	7	7	17.5%	17.5

THE BINOMIAL TEST SHOWS THAT .84 AND 1.81 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 1.248552

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
1	.2051326	1.248552	1.030544	1.522469

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H
4	2.621451	6.167871

GOODNESS OF FIT PROBABILITY

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 = 1.209431  
 95 PERCENT CONFIDENCE LIMITS = -.7487458 AND 3.167607

LC50 = 1.449643  
 95 PERCENT CONFIDENCE LIMITS = 0 AND +INFINITY

LC10 = 1291756  
 95 PERCENT CONFIDENCE LIMITS = 0 AND .6868556

\*\*\*\*\*

TITLE: Phorate Mean Length 21 Day in mm.

FILE: phordm.lng

TRANSFORM: NO TRANSFORM

NUMBER OF GROUPS: 7

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GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	Cont	1	4.1000	4.1000
1	Cont	2	3.8000	3.8000
1	Cont	3	3.6000	3.6000
1	Cont	4	3.6000	3.6000
1	Cont	5	3.7000	3.7000
1	Cont	6	4.0000	4.0000
1	Cont	7	3.8000	3.8000
1	Cont	8	3.9000	3.9000
1	Cont	9	3.8000	3.8000
1	Cont	10	4.1000	4.1000
1	Cont	11	3.7000	3.7000
1	Cont	12	3.8000	3.8000
1	Cont	13	3.8000	3.8000
1	Cont	14	3.9000	3.9000
1	Cont	15	3.8000	3.8000
1	Cont	16	3.8000	3.8000
1	Cont	17	4.2000	4.2000
1	Cont	18	4.1000	4.1000
1	Cont	19	4.4000	4.4000
1	Cont	20	4.0000	4.0000
1	Cont	21	3.7000	3.7000
1	Cont	22	3.6000	3.6000
1	Cont	23	3.5000	3.5000
1	Cont	24	3.7000	3.7000
1	Cont	25	3.9000	3.9000
1	Cont	26	4.2000	4.2000
1	Cont	27	3.9000	3.9000
1	Cont	28	3.4000	3.4000
1	Cont	29	3.3000	3.3000
1	Cont	30	3.8000	3.8000
1	Cont	31	3.7000	3.7000
1	Cont	32	3.4000	3.4000
1	Cont	33	4.3000	4.3000
2	Solv Cont	1	3.9000	3.9000
2	Solv Cont	2	3.5000	3.5000
2	Solv Cont	3	4.1000	4.1000
2	Solv Cont	4	3.6000	3.6000
2	Solv Cont	5	3.7000	3.7000
2	Solv Cont	6	3.8000	3.8000
2	Solv Cont	7	4.0000	4.0000
2	Solv Cont	8	3.6000	3.6000
2	Solv Cont	9	3.8000	3.8000
2	Solv Cont	10	4.0000	4.0000
2	Solv Cont	11	3.7000	3.7000
2	Solv Cont	12	3.9000	3.9000
2	Solv Cont	13	3.6000	3.6000
2	Solv Cont	14	3.8000	3.8000
2	Solv Cont	15	4.0000	4.0000
2	Solv Cont	16	3.9000	3.9000



2	Solv Cont	17	4.3000	4.3000
2	Solv Cont	18	4.1000	4.1000
2	Solv Cont	19	4.2000	4.2000
2	Solv Cont	20	4.0000	4.0000
2	Solv Cont	21	3.8000	3.8000
2	Solv Cont	22	3.7000	3.7000
2	Solv Cont	23	4.0000	4.0000
2	Solv Cont	24	3.7000	3.7000
2	Solv Cont	25	3.8000	3.8000
2	Solv Cont	26	3.8000	3.8000
2	Solv Cont	27	4.0000	4.0000
2	Solv Cont	28	3.6000	3.6000
2	Solv Cont	29	3.7000	3.7000
2	Solv Cont	30	3.8000	3.8000
2	Solv Cont	31	3.8000	3.8000
2	Solv Cont	32	4.0000	4.0000
3	.09	1	3.6000	3.6000
3	.09	2	3.7000	3.7000
3	.09	3	4.1000	4.1000
3	.09	4	4.1000	4.1000
3	.09	5	3.7000	3.7000
3	.09	6	3.9000	3.9000
3	.09	7	4.0000	4.0000
3	.09	8	3.7000	3.7000
3	.09	9	4.2000	4.2000
3	.09	10	4.1000	4.1000
3	.09	11	4.0000	4.0000
3	.09	12	4.1000	4.1000
3	.09	13	4.1000	4.1000
3	.09	14	3.8000	3.8000
3	.09	15	3.8000	3.8000
3	.09	16	3.7000	3.7000
3	.09	17	3.6000	3.6000
3	.09	18	3.7000	3.7000
3	.09	19	3.8000	3.8000
3	.09	20	3.7000	3.7000
3	.09	21	3.6000	3.6000
3	.09	22	4.0000	4.0000
3	.09	23	4.0000	4.0000
3	.09	24	3.5000	3.5000
3	.09	25	3.7000	3.7000
3	.09	26	4.0000	4.0000
3	.09	27	4.1000	4.1000
3	.09	28	3.8000	3.8000
3	.09	29	3.7000	3.7000
3	.09	30	3.5000	3.5000
3	.09	31	3.7000	3.7000
3	.09	32	3.8000	3.8000
3	.09	33	3.7000	3.7000
4	.21	1	3.6000	3.6000
4	.21	2	3.7000	3.7000
4	.21	3	3.9000	3.9000
4	.21	4	4.3000	4.3000
4	.21	5	3.6000	3.6000
4	.21	6	3.7000	3.7000
4	.21	7	3.8000	3.8000
4	.21	8	4.1000	4.1000
4	.21	9	4.0000	4.0000
4	.21	10	3.9000	3.9000
4	.21	11	3.9000	3.9000

4	.21	12	3.8000	3.8000
4	.21	13	4.0000	4.0000
4	.21	14	3.7000	3.7000
4	.21	15	3.8000	3.8000
4	.21	16	3.9000	3.9000
4	.21	17	3.7000	3.7000
4	.21	18	4.1000	4.1000
4	.21	19	3.6000	3.6000
4	.21	20	4.0000	4.0000
4	.21	21	3.9000	3.9000
4	.21	22	3.8000	3.8000
4	.21	23	3.9000	3.9000
4	.21	24	3.8000	3.8000
4	.21	25	4.0000	4.0000
4	.21	26	3.9000	3.9000
4	.21	27	3.9000	3.9000
4	.21	28	3.8000	3.8000
4	.21	29	3.9000	3.9000
4	.21	30	3.9000	3.9000
4	.21	31	3.8000	3.8000
4	.21	32	3.9000	3.9000
4	.21	33	4.0000	4.0000
4	.21	34	3.7000	3.7000
4	.21	35	4.0000	4.0000
4	.21	36	3.7000	3.7000
5	.41	1	3.7000	3.7000
5	.41	2	3.4000	3.4000
5	.41	3	3.6000	3.6000
5	.41	4	4.0000	4.0000
5	.41	5	3.7000	3.7000
5	.41	6	4.3000	4.3000
5	.41	7	3.7000	3.7000
5	.41	8	3.9000	3.9000
5	.41	9	3.4000	3.4000
5	.41	10	3.7000	3.7000
5	.41	11	3.5000	3.5000
5	.41	12	3.7000	3.7000
5	.41	13	3.7000	3.7000
5	.41	14	3.5000	3.5000
5	.41	15	3.7000	3.7000
5	.41	16	4.1000	4.1000
5	.41	17	4.1000	4.1000
5	.41	18	4.2000	4.2000
5	.41	19	4.0000	4.0000
5	.41	20	3.6000	3.6000
5	.41	21	3.9000	3.9000
5	.41	22	3.7000	3.7000
5	.41	23	4.3000	4.3000
5	.41	24	3.8000	3.8000
5	.41	25	3.6000	3.6000
5	.41	26	3.8000	3.8000
5	.41	27	3.7000	3.7000
6	.84	1	3.6000	3.6000
6	.84	2	4.2000	4.2000
6	.84	3	3.9000	3.9000
6	.84	4	3.9000	3.9000
6	.84	5	3.9000	3.9000
6	.84	6	3.8000	3.8000
6	.84	7	3.7000	3.7000
6	.84	8	3.7000	3.7000

6	.84	9	3.6000	3.6000
6	.84	10	3.6000	3.6000
6	.84	11	3.7000	3.7000
6	.84	12	3.6000	3.6000
6	.84	13	3.7000	3.7000
6	.84	14	3.6000	3.6000
6	.84	15	3.7000	3.7000
6	.84	16	3.7000	3.7000
6	.84	17	3.7000	3.7000
6	.84	18	3.5000	3.5000
7	1.81	1	0.0100	0.0100
7	1.81	2	3.0000	3.0000

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Phorate Mean Length 21 Day in mm.  
 File: phordm.lng Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 1 of 2

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GRP	IDENTIFICATION	N	MIN	MAX	MEAN
1	Cont	33	3.300	4.400	3.827
2	Solv Cont	32	3.500	4.300	3.850
3	.09	33	3.500	4.200	3.833
4	.21	36	3.600	4.300	3.861
5	.41	27	3.400	4.300	3.789
6	.84	18	3.500	4.200	3.728
7	1.81	2	0.010	3.000	1.505

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Phorate Mean Length 21 Day in mm.  
 File: phordm.lng Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 2 of 2

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GRP	IDENTIFICATION	VARIANCE	SD	SEM
1	Cont	0.066	0.258	0.045
2	Solv Cont	0.036	0.190	0.034
3	.09	0.040	0.199	0.035
4	.21	0.024	0.154	0.026
5	.41	0.063	0.252	0.048
6	.84	0.027	0.164	0.039
7	1.81	4.470	2.114	1.495

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Phorate Mean Length 21 Day in mm.  
 File: phordm.lng Transform: NO TRANSFORM

ANOVA TABLE

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SOURCE	DF	SS	MS	F
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Between	6	10.906	1.818	26.735
Within (Error)	174	11.917	0.068	
Total	180	22.823		

Critical F value = 2.18 (0.05,6,120)  
 Since F > Critical F REJECT Ho:All groups equal

Phorate Mean Length 21 Day in mm.  
 File: phordm.lng Transform: NO TRANSFORM

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	Cont	3.827	3.827		
2	Solv Cont	3.850	3.850	-0.351	
3	.09	3.833	3.833	-0.094	
4	.21	3.861	3.861	-0.538	
5	.41	3.789	3.789	0.567	
6	.84	3.728	3.728	1.302	
7	1.81	1.505	1.505	12.229	*

Bonferroni T table value = 2.43 (1 Tailed Value, P=0.05, df=120,6)

Phorate Mean Length 21 Day in mm.  
 File: phordm.lng Transform: NO TRANSFORM

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	Cont	33			
2	Solv Cont	32	0.157	4.1	-0.023
3	.09	33	0.156	4.1	-0.006
4	.21	36	0.153	4.0	-0.034
5	.41	27	0.164	4.3	0.038
6	.84	18	0.186	4.8	0.099
7	1.81	2	0.461	12.1	2.322

Phorate Mean Length 21 Day in mm.  
 File: phordm.lng Transform: NO TRANSFORM

ANOVA TABLE

SOURCE	DF	SS	MS	F
--------	----	----	----	---

Between	6	10.906	1.818	26.735
Within (Error)	174	11.917	0.068	
Total	180	22.823		

Critical F value = 2.18 (0.05,6,120)  
 Since F > Critical F REJECT Ho:All groups equal

Phorate Mean Length 21 Day in mm.  
 File: phordm.lng Transform: NO TRANSFORM

TUKEY method of multiple comparisons

GROUP	IDENTIFICATION	TRANSFORMED MEAN	ORIGINAL MEAN	GROUP									
				0	0	0	0	0	0	0	0		
7	1.81	1.505	1.505	\									
6	.84	3.728	3.728	*	\								
5	.41	3.789	3.789	*	.	\							
1	Cont	3.827	3.827	*	.	.	\						
3	.09	3.833	3.833	*	.	.	.	\					
2	Solv Cont	3.850	3.850	*	.	.	.	.	\				
4	.21	3.861	3.861	*	.	.	.	.	.	\			

\* = significant difference (p=0.05) . = no significant difference  
 Tukey value (7,174) = 4.24 s = 0.068

Phorate Mean Length 21 Day in mm.  
 File: phordm.lng Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	Cont	33	3.827	3.827	3.843
2	Solv Cont	32	3.850	3.850	3.843
3	.09	33	3.833	3.833	3.843
4	.21	36	3.861	3.861	3.843
5	.41	27	3.789	3.789	3.789
6	.84	18	3.728	3.728	3.728
7	1.81	2	1.505	1.505	1.505

Phorate Mean Length 21 Day in mm.  
 File: phordm.lng Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
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Cont	3.843				
Solv Cont	3.843	0.247		1.66	k= 1, v=174
.09	3.843	0.249		1.73	k= 2, v=174
.21	3.843	0.254		1.75	k= 3, v=174
.41	3.789	0.565		1.77	k= 4, v=174
.84	3.728	1.297		1.77	k= 5, v=174
1.81	1.505	12.185	*	1.78	k= 6, v=174

s = 0.262

Note: df used for table values are approximate when  $v > 20$ .

Daphnia 21 Day Weight/Daphnid in Mg. Phorate  
 File: PhoraDM.wt Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 1 of 2

GRP	IDENTIFICATION	N	MIN	MAX	MEAN
1	Controls	4	0.375	0.456	0.411
2	Solvent Contrl	4	0.390	0.535	0.444
3	0.09 ug/L	4	0.425	0.500	0.455
4	0.21 ug/L	4	0.410	0.587	0.485
5	0.41 ug/L	4	0.400	0.538	0.464
6	0.84 ug/L	4	0.160	0.400	0.323
7	1.81 ug/L	2	0.000	0.300	0.150

Daphnia 21 Day Weight/Daphnid in Mg. Phorate  
 File: PhoraDM.wt Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 2 of 2

GRP	IDENTIFICATION	VARIANCE	SD	SEM
1	Controls	0.001	0.034	0.017
2	Solvent Contrl	0.004	0.064	0.032
3	0.09 ug/L	0.001	0.034	0.017
4	0.21 ug/L	0.006	0.075	0.038
5	0.41 ug/L	0.004	0.066	0.033
6	0.84 ug/L	0.013	0.113	0.057
7	1.81 ug/L	0.045	0.212	0.150

Daphnia 21 Day Weight/Daphnid in Mg. Phorate  
 File: PhoraDM.wt Transform: NO TRANSFORM

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	6	0.212	0.035	5.000
Within (Error)	19	0.133	0.007	
Total	25	0.345		

Critical F value = 2.63 (0.05,6,19)  
 Since F > Critical F REJECT Ho:All groups equal

Daphnia 21 Day Weight/Daphnid in Mg. Phorate

File: PhoraDM.wt

Transform: NO TRANSFORM

## BONFERRONI T-TEST - TABLE 1 OF 2

Ho:Control&lt;Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	Controls	0.411	0.411		
2	Solvent Contrl	0.444	0.444	-0.559	
3	0.09 ug/L	0.455	0.455	-0.749	
4	0.21 ug/L	0.485	0.485	-1.261	
5	0.41 ug/L	0.464	0.464	-0.890	
6	0.84 ug/L	0.323	0.323	1.480	
7	1.81 ug/L	0.150	0.150	3.600	*

Bonferroni T table value = 2.63 (1 Tailed Value, P=0.05, df=19,6)

Daphnia 21 Day Weight/Daphnid in Mg. Phorate

File: PhoraDM.wt

Transform: NO TRANSFORM

## BONFERRONI T-TEST - TABLE 2 OF 2

Ho:Control&lt;Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	Controls	4			
2	Solvent Contrl	4	0.155	37.8	-0.033
3	0.09 ug/L	4	0.155	37.8	-0.044
4	0.21 ug/L	4	0.155	37.8	-0.075
5	0.41 ug/L	4	0.155	37.8	-0.053
6	0.84 ug/L	4	0.155	37.8	0.088
7	1.81 ug/L	2	0.190	46.3	0.261

Daphnia 21 Day Weight/Daphnid in Mg. Phorate

File: PhoraDM.wt

Transform: NO TRANSFORM

## ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	6	0.212	0.035	5.000
Within (Error)	19	0.133	0.007	
Total	25	0.345		

Critical F value = 2.63 (0.05,6,19)

Since F &gt; Critical F REJECT Ho:All groups equal

Daphnia 21 Day Weight/Daphnid in Mg. Phorate

File: PhoraDM.wt

Transform: NO TRANSFORM



TUKEY method of multiple comparisons

GROUP	IDENTIFICATION	TRANSFORMED MEAN	ORIGINAL MEAN	GROUP							
				0	0	0	0	0	0	0	0
				7	6	1	2	3	5	4	
7	1.81 ug/L	0.150	0.150	\							
6	0.84 ug/L	0.323	0.323	.	\						
1	Controls	0.411	0.411	*	.	\					
2	Solvent Contrl	0.444	0.444	*	.	.	\				
3	0.09 ug/L	0.455	0.455	*	.	.	.	\			
5	0.41 ug/L	0.464	0.464	*	.	.	.	.	\		
4	0.21 ug/L	0.485	0.485	*	.	.	.	.	.	\	

\* = significant difference (p=0.05)

. = no significant difference

Tukey value (7,19) = 4.65

s = 0.007

Daphnia 21 Day Weight/Daphnid in Mg. Phorate  
File: PhoraDM.wt Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	Controls	4	0.411	0.411	0.452
2	Solvent Contrl	4	0.444	0.444	0.452
3	0.09 ug/L	4	0.455	0.455	0.452
4	0.21 ug/L	4	0.485	0.485	0.452
5	0.41 ug/L	4	0.464	0.464	0.452
6	0.84 ug/L	4	0.323	0.323	0.323
7	1.81 ug/L	2	0.150	0.150	0.150

Daphnia 21 Day Weight/Daphnid in Mg. Phorate  
File: PhoraDM.wt Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
Controls	0.452				
Solvent Contrl	0.452	0.692		1.73	k= 1, v=19
0.09 ug/L	0.452	0.692		1.81	k= 2, v=19
0.21 ug/L	0.452	0.692		1.84	k= 3, v=19
0.41 ug/L	0.452	0.692		1.85	k= 4, v=19
0.84 ug/L	0.323	1.481		1.86	k= 5, v=19
1.81 ug/L	0.150	3.603	*	1.87	k= 6, v=19

s = 0.084

Note: df used for table values are approximate when v > 20.

TITLE: Phorate Total Offspring/Daphnid (mean NUMBER/reproductive day)  
 FILE: phortot.off  
 TRANSFORM: NO TRANSFORM NUMBER OF GROUPS: 7

GRP	IDENTIFICATION	REP	VALUE	TRANS VALUE
1	Control	1	47.7000	47.7000
1	Control	2	48.7000	48.7000
1	Control	3	49.7800	49.7800
1	Control	4	50.3000	50.3000
2	Solv. Cont.	1	60.6000	60.6000
2	Solv. Cont.	2	47.6000	47.6000
2	Solv. Cont.	3	63.6000	63.6000
2	Solv. Cont.	4	48.2000	48.2000
3	.09	1	53.0000	53.0000
3	.09	2	48.4000	48.4000
3	.09	3	42.3000	42.3000
3	.09	4	35.1000	35.1000
4	.21	1	42.1000	42.1000
4	.21	2	48.8000	48.8000
4	.21	3	49.1000	49.1000
4	.21	4	55.9000	55.9000
5	.41	1	54.0000	54.0000
5	.41	2	37.0000	37.0000
5	.41	3	47.9000	47.9000
5	.41	4	51.5000	51.5000
6	.84	1	66.7000	66.7000
6	.84	2	51.3900	51.3900
6	.84	3	39.9000	39.9000
6	.84	4	48.0000	48.0000
7	1.81	1	15.4500	15.4500
7	1.81	2	19.0000	19.0000
7	1.81	3	13.3700	13.3700
7	1.81	4	18.6300	18.6300

Phorate Total Offspring/Daphnid  
 File: phortot.off Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 1 of 2

GRP	IDENTIFICATION	N	MIN	MAX	MEAN
1	Control	4	47.700	50.300	49.120
2	Solv. Cont.	4	47.600	63.600	55.000
3	.09	4	35.100	53.000	44.700
4	.21	4	42.100	55.900	48.975
5	.41	4	37.000	54.000	47.600
6	.84	4	39.900	66.700	51.497
7	1.81	4	13.370	19.000	16.613

Phorate Total Offspring/Daphnid  
 File: phortot.off Transform: NO TRANSFORM

SUMMARY STATISTICS ON TRANSFORMED DATA TABLE 2 of 2

GRP	IDENTIFICATION	VARIANCE	SD	SEM
1	Control	1.340	1.158	0.579
2	Solv. Cont.	68.773	8.293	4.146
3	.09	60.167	7.757	3.878
4	.21	31.756	5.635	2.818
5	.41	56.207	7.497	3.749
6	.84	125.954	11.223	5.611
7	1.81	7.212	2.685	1.343

Phorate Total Offspring/Daphnid  
 File: phortot.off Transform: NO TRANSFORM

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	6	3949.494	658.249	13.112
Within (Error)	21	1054.226	50.201	
Total	27	5003.720		

Critical F value = 2.57 (0.05,6,21)  
 Since  $F > \text{Critical } F$  REJECT  $H_0$ :All groups equal

Phorate Total Offspring/Daphnid  
 File: phortot.off Transform: NO TRANSFORM

BONFERRONI T-TEST - TABLE 1 OF 2  $H_0$ :Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	Control	49.120	49.120		
2	Solv. Cont.	55.000	55.000	-1.174	
3	.09	44.700	44.700	0.882	
4	.21	48.975	48.975	0.029	
5	.41	47.600	47.600	0.303	
6	.84	51.497	51.497	-0.475	
7	1.81	16.613	16.613	6.488	*

Bonferroni T table value = 2.60 (1 Tailed Value,  $P=0.05$ ,  $df=21,6$ )

Phorate Total Offspring/Daphnid

File: phbrtot.off

Transform: NO TRANSFORM

BONFERRONI T-TEST - TABLE 2 OF 2

Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	Control	4			
2	Solv. Cont.	4	13.036	26.5	-5.880
3	.09	4	13.036	26.5	4.420
4	.21	4	13.036	26.5	0.145
5	.41	4	13.036	26.5	1.520
6	.84	4	13.036	26.5	-2.377
7	1.81	4	13.036	26.5	32.508

Phorate Total Offspring/Daphnid

File: phortot.off

Transform: NO TRANSFORM

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	6	3949.494	658.249	13.112
Within (Error)	21	1054.226	50.201	
Total	27	5003.720		

Critical F value = 2.57 (0.05,6,21)

Since F > Critical F REJECT Ho:All groups equal

Phorate Total Offspring/Daphnid

File: phortot.off

Transform: NO TRANSFORM

TUKEY method of multiple comparisons

GROUP	IDENTIFICATION	TRANSFORMED MEAN	ORIGINAL MEAN	GROUP							
				7	3	5	4	1	6	2	
7	1.81	16.613	16.613	\							
3	.09	44.700	44.700	*	\						
5	.41	47.600	47.600	*	.	\					
4	.21	48.975	48.975	*	.	.	\				
1	Control	49.120	49.120	*	.	.	.	\			
6	.84	51.497	51.497	*	.	.	.	.	\		
2	Solv. Cont.	55.000	55.000	*	.	.	.	.	.	\	

\* = significant difference (p=0.05)

Tukey value (7,21) = 4.62

. = no significant difference

s = 50.201

Phorate Total Offspring/Daphnid  
 File: phortot.off Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	Control	4	49.120	49.120	52.060
2	Solv. Cont.	4	55.000	55.000	52.060
3	.09	4	44.700	44.700	48.193
4	.21	4	48.975	48.975	48.193
5	.41	4	47.600	47.600	48.193
6	.84	4	51.497	51.497	48.193
7	1.81	4	16.613	16.613	16.613

Phorate Total Offspring/Daphnid  
 File: phortot.off Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
Control	52.060				
Solv. Cont.	52.060	0.587		1.72	k= 1, v=21
.09	48.193	0.185		1.80	k= 2, v=21
.21	48.193	0.185		1.83	k= 3, v=21
.41	48.193	0.185		1.84	k= 4, v=21
.84	48.193	0.185		1.85	k= 5, v=21
1.81	16.613	6.488	*	1.85	k= 6, v=21

s = 7.085

Note: df used for table values are approximate when v > 20.

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

Dilution Controls  
ONLY

B. Montague Phorate Chronic 21 Day Daphnid

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
1.81	33	32	96.96969	0
.84	33	15	45.4545	0
.41	33	6	18.1818	0
.21	40	4	10	0
9.000001E-02		33	0	0

0

BECAUSE THE NUMBER OF ORGANISMS USED WAS SO LARGE, THE 95 PERCENT CONFIDENCE INTERVALS CALCULATED FROM THE BINOMIAL PROBABILITY ARE UNRELIABLE. USE THE INTERVALS CALCULATED BY THE OTHER TESTS.

AN APPROXIMATE LC50 FOR THIS SET OF DATA IS .8871742

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
3	5.369509E-02		.7447086	.6159563

.9288642

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
4	7.443428E-02	6.706833E-02	1

SLOPE = 3.088159  
 95 PERCENT CONFIDENCE LIMITS = 2.288402 AND 3.887917

LC50 = .7209131  
 95 PERCENT CONFIDENCE LIMITS = .595918 AND .8896428

LC10 = .2796628  
 95 PERCENT CONFIDENCE LIMITS = .1957876 AND .3556242

\*\*\*\*\*

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
3	5.369509E-02		.7447086	.6159563

.9288642

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY	
4	6.706833E-02	1	7.443428E-02	

SLOPE = 3.088159  
 95 PERCENT CONFIDENCE LIMITS = 2.288402 AND 3.887917

LC50 = .7209131  
95 PERCENT CONFIDENCE LIMITS = .595918 AND .8896428

LC10 = .2796628  
95 PERCENT CONFIDENCE LIMITS = .1957876 AND .3556242

\*\*\*\*\*

DO YOU WISH TO RUN ANOTHER DATA SET?  
ENTER Y OR N.  
?

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD  
SPAN            G            LC50            95 PERCENT CONFIDENCE LIMITS  
3                5.369509E-02            .7447086            .6159563  
.9288642

RESULTS CALCULATED USING THE PROBIT METHOD  
ITERATIONS    G            H            GOODNESS OF FIT PROBABILITY  
4                6.706833E-02            1            7.443428E-02

SLOPE        =    3.088159  
95 PERCENT CONFIDENCE LIMITS = 2.288402        AND        3.887917

LC50 = .7209131  
95 PERCENT CONFIDENCE LIMITS = .595918 AND .8896428

LC10 = .2796628  
95 PERCENT CONFIDENCE LIMITS = .1957876 AND .3556242

\*\*\*\*\*

DO YOU WISH TO RUN ANOTHER DATA SET?  
ENTER Y OR N.  
?