State of New Jersey CHRISTINE TODD WHITMAN GOVERNOR

SEDIMENT TOXICITY TEST USING THE AMPHIPOD

Hyalella azteca
Watershed Management Areas 1 and 11
(Upper Delaware River Basin)



New Jersey Department of Environmental Protection ROBERT C. SHINN, JR. COMMISSIONER

April 1999



New Jersey Department of Environmental Protection Division of Watershed Management P.O. Box 427, Trenton, NJ 08625-0427

WATER MONITORING MANAGEMENT

James Mumman, Administrator

April 1999

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Bureau of Freshwater and Biological Monitoring Biomonitoring Section

Assay Number(s): 99H002a, 99H002b, 99H002c, 99H002d

Report Prepared By:

Thomas Miller

Analysts:

Thomas Miller Dean Bryson Victor Poretti

Samplers:

Thomas Miller Dean Bryson Victor Poretti

Supervisor:

Paul Olsen

Chief

Bureau of Freshwater & Biological Monitoring Alfred Korndoerfer, Jr.

EXECUTIVE SUMMARY

Toxicity tests using the amphipod *Hyalella azteca* were performed on sediments collected from four sites in the upper Delaware River system. This initiative was undertaken by the Bureau of Freshwater and Biological Monitoring in conjunction with the Office of Watershed Management, both administered under the New Jersey Department of Environmental Protection's Division of Watershed Management. The sites tested were situated on the Jacksonburg Creek, an unnamed tributary (UNT) to Paulinskill, an unnamed tributary to the Pequest River and Plum Brook. The first three sites are within Watershed Management Area # 1, and Plum Brook is in Watershed Management Area # 11. All sites are within the Upper Delaware "Water Region". Suspected toxicity at three of the sites (UNT to Paulinskill, UNT to Pequest River and Plum Brook) was based on their "moderately impaired" biological assessments (i.e. degraded quality of benthic macroinvertebrate communities) as compared to their "non-impaired" assessment found in previous survey(s) of New Jerseys statewide Ambient Biomonitoring Network (AMNET). A reference site was selected at Jacksonburg Creek because of its "non-impaired" AMNET assessment. The sediment toxicity tests were conducted to evaluate whether or not toxic materials in the streambed sediments could account for the observed macroinvertebrate impairment. Tests were conducted in accordance with the Bureau Standard Operating Procedures, which incorporate protocols recognized by the U.S. Environmental Protection Agency. When test results were statistically compared to that of the reference, the test sites did not exhibit acute toxicity, as measured by survival, or chronic toxicity as measured by growth, of test organisms.

INTRODUCTION

The Ambient Biomonitoring Network (AMNET) program of the New Jersey Department of Environmental Protection (NJDEP), Bureau of Freshwater and Biological Monitoring (BFBM), is designed to establish a biological database for use in gauging stream quality throughout the state. This database, in turn, can be an invaluable aid to New Jerseys water quality and watershed planning and management efforts. Levels of impairment are shown through the use of Rapid Bioassessment Protocols (RBP) advised by the U.S. Environmental Protection Agency (EPA)(1). The RBP assesses impairment through the collection, identification, categorizing, and quantification of instream macroinvertebrate communities. Although the RBP is an excellent way in which to assess impairment, it may sometimes be difficult to distinguish whether impairment is due to water quality degradation or habitat destruction.

Sediment toxicity testing is an additional tool used to determine whether toxicity is the cause of impairment, before resorting to costly chemical monitoring. The test organism, *Hyalella azteca* is an epibenthic detritivore, reported to also digest bacteria and algae from ingested sediment particles (2). This amphipod crustacean inhabits lakes, ponds, and streams throughout North and South America, typically burrowing into the sediment surface (3,4). *H. azteca* is a sensitive "benchmark" species, which can be cultured in the laboratory with relative ease.

In March of 1999, the Bureau of Freshwater and Biological Monitoring conducted sediment toxicity tests on four stream sites, within an area of northwestern New Jersey, which had exhibited varying degrees of impairment in previous AMNET sampling (5,10). The new initiative was designed to support management efforts in Watershed Management Areas (WMA) #1 and 11. Administratively, this includes the Upper Delaware river drainage, in the Upper Delaware "Water Region".

METHODS

Sample sites were selected based on previous AMNET results (5, 10) (see appendix A), proximity to urban and/or agricultural areas, and proximity of point source discharges (i.e. effluents from facilities with New Jersey Pollutant Discharge Elimination System (NJPDES) permits). The sites selected are as follows (see maps):

AMNET	BIOLOGICAL	
STATION#	<u>ASSESSMENT</u>	<u>LOCATION</u> (see maps)
AN0028	non-impaired	Jacksonburg Ck @ Rt.602, Hardwick Twp.
AN0016	moderately impaired	UNT to Paulinskill @ Meadows Rd., Lafayette
		Twp.
AN0036	moderately impaired	UNT to Pequest River @ Rt. 603, Green Twp.
AN0093	moderately impaired	Plum Brook @ Pine Hill Rd., Delaware Twp.

Sediment samples were collected from all sites on March 10, 1999. At each station the sediment was collected in the stream channel using a stainless steel scoop sampler and placed into one liter amber glass bottles, then stored at # 4EC until the start of the test (6).

Prior to test initiation the sample sites were assigned assay numbers, in accordance with our ongoing series of toxicity tests, as follows:

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99H002a = AN0028 (reference, nonimpaired site)

99H002b = AN0016

99H002c = AN0036

99H002d = AN0093
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Testing methodology followed the BFBM Standard Operating Procedures (7). 24 hours prior to the start of the test, the sediment from each station was mixed to provide a homogeneous sample, and hand picked of any visible indigenous organisms. For each site, 100 ml of sediment was added to each of the five 300 ml replicate test vessels and topped with laboratory grade freshwater to the 250 ml mark. The test vessels were then held at the test temperature (23EC) for 24 hours to allow the sediment to settle (7). After this time period, the overlying water was siphoned, and fresh water was added. A control set of replicates was also set up using 250 ml of laboratory grade freshwater only.

One to seven-day old *H. azteca* juveniles were collected, from our cultures, and held for one week prior to the start of the test (7).

Testing was initiated on March 16, 1999 at 10:40 hours, by adding ten 7 to 14- day old organisms from the holding chamber to each test replicate series. Each day the overlying water was exchanged, and each test replicate was fed 1.5 ml of yeast, CEROPHYLL7, Trout chow (YCT)(8), and 1.5 ml of the green algae *Selenastrum capricornutum* at a concentration of 35 X 10⁶ cells ml⁻¹ (after centrifugation). Mortalities were noted if visible; pH, dissolved oxygen, and conductivity were measured from aliquots of each test series; measurements were made at the start of the test and after each 24 hour period (see Table 1).

The test was concluded after ten days (March 26, 1999). As a measure of acute toxicity, live organisms were counted against those dead or missing (see Table 2). As an indication of chronic toxicity, dry weights of surviving organisms were measured (see Table 3). Statistical analysis was performed following EPA guidelines (8). Results of the reference test were compared against the control, and, providing the reference and the control were statistically similar, the remaining tests were compared to the reference.

RESULTS

The tests were valid by virtue of meeting the acceptability requirements of \$80% survival (see Table 2) in the control test series (7). The survival data was not distributed normally as analyzed by the Shapiro-Wilks test for normality, therefore, the Wilcoxan Rank Sum Test was used when comparing test survival results. There was no significant difference in survival results between the reference test (99H002a) and the control. All test samples were then compared to the reference. The survival data was not distributed normally as analyzed by the Shapiro-Wilks test for normality, therefore, the Wilcoxan Rank Sum Test was used when comparing test survival results. The results for tests 99H002b, 99H002c and 99H002d all showed no significant difference in mortality from that of the reference sample. Dry weights of these test samples were then compared to that of the reference (see Table 3). The dry weight data for 99H002b was distributed

normally as analyzed by the Shapiro-Wilks test, and therefore, an F-Test and T-Test was used when comparing test dry weight results. In this comparison the test samples showed no significant difference from the reference (see appendix B for statistical printout). The dry weight data for 99H002c and 99H002d was not distributed normally, as analyzed by the Shapiro-Wilks test for normality, therefore, the Wilcoxan Rank Sum Test was used when comparing test dry weight analysis for these tests. The results for tests 99H002c and 99H002d showed no significant difference in growth from that of the reference sample.

Although all indigenous organisms observed in the samples before the start of the test were removed, some did remain; however, their presence did not invalidate test results. Test chambers 99H002a and 99H002b contained several *chironomid*, or midge, larvae. Test chamber 99H002b also contained a caddisfly, isopod, watersnipe and a beetle larva. Test chamber 99H002c contained an amphipod and a snail. Test chamber 99H002d contained a beetle and a dobsonfly larva.

DISCUSSION

The test sites (one each on UNT to Paulinskill, UNT to Pequest River and Plum Brook) in WMA's 1 and 11 were chosen based on the results of previous macroinvertebrate studies and the presence of likely sources of impairment; these included the proximity of NJPDES facilities, and of urbanization, or agriculture. The reference site at Jacksonburg Creek, was chosen because of its prior "non-impaired" bioassessment in the AMNET survey (5, 10), and because it is within the same Water Region (Upper Delaware River Basin) as the test sites. Also considered in choosing the reference site, were similarities in stream morphology and position within the New Jersey Ecomap or ecoregion scheme (1,9).

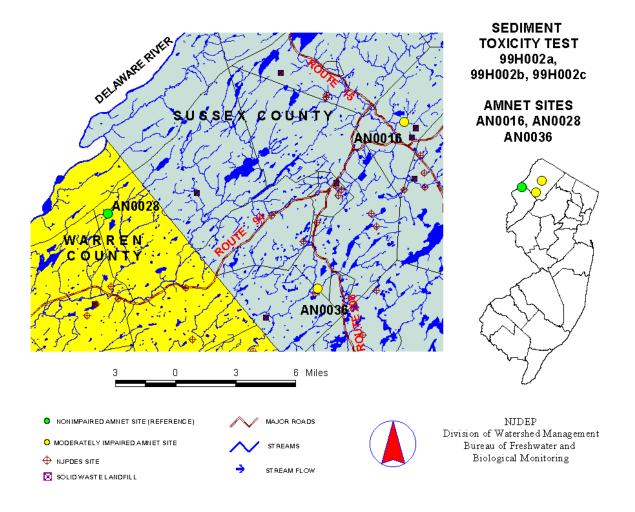
Sites AN0016 (UNT to Paulinskill), AN0036 (UNT to Pequest River) and AN0093 (Plum Brook), were chosen for testing based on degraded bioassessment results (from "non-impaired" to "moderately impaired") (5,10), and the high agricultural land use in the area which may have influenced the bioassessment results. There are also a few landfills and NJPDES sites upstream of the sampling points, including Hamm's Landfill Expansion, Sussex County Landfill, Andover Township MUA, Long Pond School and DRG Medical Packaging Inc.

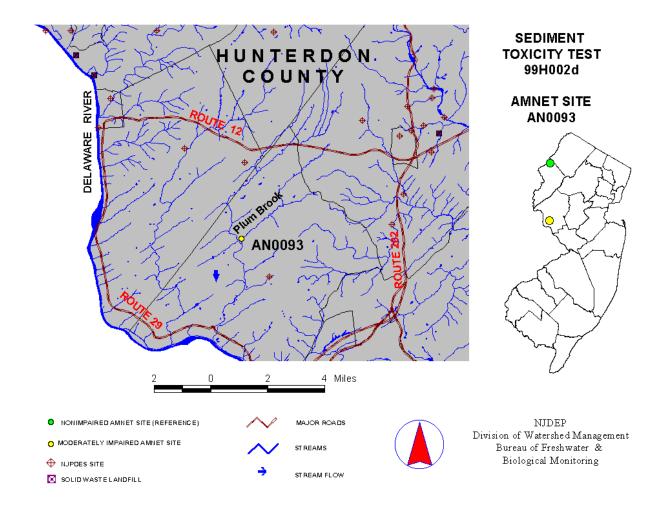
Survival and dry weight results showed no significant differences between the reference and the test site treatments. Since the test site results did not indicate acute toxicity, the reduced impairment levels found may have been due to other factors including habitat degradation and/or various physiochemical parameters (e.g. temperature, turbidity, low dissolved oxygen, pH, etc...). Impairment may also have been caused by the presence of other toxic substances at chronically, but not acutely, toxic levels, which could be introduced episodically, rather than continuously, into the stream. Therefore, it is advisable by these study results, that supplemental sampling be performed for target analytes, such as excessive nutrients (usually forms of nitrogen or phosphorus), and pesticides or other known toxic compounds.

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MAPS





Bureau of Freshwater and Biological Monitoring Sediment Toxicity Tests Watershed Management Areas # 1 and 11 Upper Delaware River Basin

Table 1 Test Chamber Chemical/Physical Parameters

	. DEV. % CV
nH 79 73 75 0	17 225
pri 7.5 7.5 0	0.17 2.25
cond. Fmhos 150 141 146 3	2.20
D.O. mg/L 7.9 7.0 7.6 0	0.35 4.56
99H002a HIGH LOW AVG. STD	. DEV. % CV
pH 7.7 7.1 7.5 0	3.24
cond. Fmhos 170 134 159 10	0.11 6.34
D.O. mg/L 8.1 6.1 7.5 0	0.60 8.00
99H002b HIGH LOW AVG. STD	. DEV. % CV
pH 7.9 7.3 7.7 0	2.83
cond. Fmhos 237 202 217 13	1.96 5.52
D.O. mg/L 8.0 5.5 7.0 0	0.95 13.66
99H002c HIGH LOW AVG. STD	. DEV. % CV
pH 8.4 7.7 7.9 0	2.83
cond. Fmhos 457 272 320 52	2.90 16.54
D.O. mg/L 7.8 5.6 6.7 0	0.91 13.64
99H002d HIGH LOW AVG. STD	. DEV. % CV
pH 8.4 7.7 7.9 0	0.22 2.83
cond. F mhos 457 272 320 52	2.90 16.54
D.O. mg/L 7.8 5.6 6.7 0	0.91 13.64

Bureau of Freshwater and Biological Monitoring

Sediment Toxicity Tests Watershed Management Areas # 1 and 11 Upper Delaware River Basin

TABLE 2

MORTALITY DATA (number surviving)

ASSAY#	REP. A	REP. B	REP. C	REP. D	REP. E	%survival
Control	10	10	10	10	10	100
99H002a	9	9	10	10	10	96
99H002b	10	7	9	10	9	90
99Н002с	9	10	10	10	10	98
99H002d	10	8	8	9	9	88

Statistical Analysis

Test Endpoint: Survival

Test Used: Wilcoxan Rank Sum Test

Results: 99H002a: no significant difference from control

99H002b: no significant difference from reference station no significant difference from reference station 99H002d: no significant difference from reference station

^{*}see appendix B for statistical printout

Bureau of Freshwater and Biological Monitoring Sediment Toxicity Tests Watershed Management Areas # 1 and 11 Upper Delaware River Basin

TABLE 3WEIGHT DETERMINATION

Drying Oven Temperature: 105EC Duration: 17.5 hours Analyst: T. Miller

REPLICATE.		WGT. OF BOAT (mg)	DRY WGT: BOAT + ORGANISMS (mg)	TOTAL WGT. OF ORGANISMS (mg)	NUMBER OF ORGANISMS	ORGANISM AVG. DRY WGT. (mg)	GROUP AVG. (mg)
CONTROL	A	431.66	432.64	0.98	10	0.098	
	В	435.95	437.01	1.06	10	0.108	
	С	434.68	435.98	1.30	10	0.130	0.108
	D	432.53	433.62	1.09	10	0.109	
	Е	433.66	434.65	0.99	10	0.099	
99H002a	A	436.71	438.29	1.58	9	0.176	
	В	433.60	435.18	1.58	9	0.176	
	С	435.31	437.08	1.77	10	0.177	0.175
	D	432.54	434.30	1.76	10	0.176	
	Е	435.06	436.78	1.72	10	0.172	
99H002b	A	432.09	433.80	1.71	10	0.171	
	В	435.11	436.17	1.06	7	0.151]
	С	433.76	435.63	1.87	9	0.208	0.159
	D	433.58	434.78	1.20	10	0.120	
	Е	433.09	434.39	1.30	9	0.144	
99H002c	A	435.29	437.07	1.78	9	0.198	
	В	436.81	438.78	1.97	10	0.197]
	С	433.38	435.38	2.00	10	0.200	0.196
	D	431.12	433.02	1.90	10	0.190]
	Е	436.90	438.86	1.96	10	0.196]
99H002d	A	435.78	437.22	1.44	10	0.144	
	В	433.72	435.73	2.01	8	0.251	1
	С	435.89	437.38	1.49	8	0.186	0.193
	D	435.73	437.35	1.62	9	0.180]
	Е	434.16	435.98	1.82	9	0.202	1

Bureau of Freshwater and Biological Monitoring Sediment Toxicity Tests Watershed Management Areas # 1 and 11 Upper Delaware River Basin

WEIGHT DETERMINATION continued...

Statistical Analysis Test Endpoint: Growth

Results:

Wilcoxan Rank Sum Test 99H002a: no significant difference from control no significant difference from reference station wilcoxan Rank Sum Test 99H002c: no significant difference from reference station no significant difference from reference station no significant difference from reference station no significant difference from reference station

^{*}see appendix B for statistical printout

Bureau of Freshwater and Biological Monitoring

APPENDIX A

AMNET DATA(5)

Sediment Toxicity Tests
Watershed Management Areas # 1 and 11
Upper Delaware River Basin

Upper Delaware Basin - Newton E USGS Quadrangle

Station: AN0016

Paulinskill Trib, Lafayette Meadows Rd, Lafayette Twp, Sussex County

Date Sampled: 10/16/97

	Family Tolerance	Number of	
Family	Value (FTV)	Individuals	
Gammaridae	4	50	
Sphaeriidae	8	11	
Chironomidae	6	10	
Elmidae	4	8	
Tubificidae	10	5	
Asellidae	8	4	
Psephenidae	4	3	
Hydropsychidae	4	2	
BloodRed Chironomidae	8	2	
Planorbidae	6	1	
Lepidostomatidae	1	1	
Ceratopogonidae	6	1	
Limnephilidae	4	1	
Simulidae	6	1	

Statistical Analysis

Number of Taxa: 14

Total Number of Individuals: 100

% Contribution of Dominant Family: 50.00 % (Gammaridae)

Family Biotic Index: 5.21

Scraper/Filterer Collector Ratio: 0.50

Shredder/Total Ratio: 0.06

E+P+T (Ephemeroptera, Plecoptera, Trichoptera): 3

% EPT: 4.00 EPT/C: 0.33 NJIS Rating: 15

Biological Condition: Moderately Impaired

Habitat Analysis: 147

Observations

Streamwater: Clear....Flow: Slow....Width/Depth (ft): 7/1

Substrate: Cobbles,gravel....StreamBank Vegetation/Stability: Trees,shrubs/Stable

Canopy: Mostly Closed....Other: Forested/Agricultural; Water temp.11.4 /pH 7.8 /DO 6.8 /Cond.853

Upper Delaware Basin - Flatbrookville USGS Quadrangle

Station: AN0028

Jacksonburg Ck, Rt 602, Hardwick Twp, Warren County

Date Sampled: 11/13/97

	Family Tolerance	Number of	
Family	Value (FTV)	Individuals	
Taeniopterygidae	2	13	
Chironomidae	6	12	
Leptophlebiidae	2	10	
Sphaeriidae	8	9	
Brachycentridae	1	7	
Tubificidae	10	6	
Hydropsychidae	4	5	
Sialidae	4	5	
Elmidae	4	4	
Protoneuridae	9	3	
Aeshnidae	3	3	
Polycentropodidae	6	3	
Philopotamidae	3	2	
Ephemerellidae	1	2	
BloodRed Chironomidae	8	2	
Odontoceridae	0	2	
Rhyacophilidae	0	2	
Heptageniidae	4	2	
Perlidae	1	1	
Calopterygidae	5	1	
Cordulegastridae	3	1	
Planariidae	4	1	
Oligoneuriidae	2	1	
Lumbricidae	10	1	
Molannidae	6	1	
Pteronarcidae	0	1	

Statistical Analysis

Number of Taxa: 26

Total Number of Individuals: 100

% Contribution of Dominant Family: 13.00 % (Taeniopterygidae)

Family Biotic Index: 4.30

Scraper/Filterer Collector Ratio: 0.55

Shredder/Total Ratio: 0.21

E+P+T (Ephemeroptera, Plecoptera, Trichoptera): 14

% EPT: 52.00 EPT/C: 3.71 NJIS Rating: 30

Biological Condition: Nonimpaired

Habitat Analysis: 182

Observations

Streamwater: Clear....Flow: Moderate....Width/Depth (ft): 10/1

Substrate: Cobbles,gravel....StreamBank Vegetation/Stability: Trees/Stable

Canopy: Mostly Open....Other: Forested/Suburban; Water temp.4.7 /pH 6.7 /DO - /Cond.57

Upper Delaware Basin - Tranquility USGS Quadrangle

Station: AN0036

Pequest R Trib, Brighton Rd, Brighton, Sussex County

Date Sampled: 09/25/97

	Family Tolerance	Number of	
Family	Value (FTV)	Individuals	
Gammaridae	4	69	
Physidae	7	9	
Hydrobiidae	8	8	
Elmidae	4	5	
Corixidae	9	4	
Blood Red Chironomidae	8	1	
Helicopsychidae	3	1	
Protoneuridae	9	1	
Tubificidae	10	1	
Sphaeriidae	8	1	

Statistical Analysis

Number of Taxa: 10

Total Number of Individuals: 100

% Contribution of Dominant Family: 69.00 % (Gammaridae)

Family Biotic Index: 4.97

Scraper/Filterer Collector Ratio: 6.00

Shredder/Total Ratio: 0.00

E+P+T (Ephemeroptera, Plecoptera, Trichoptera): 1

% EPT: 1.00 EPT/C: 1.00 NJIS Rating: 9

Biological Condition: Moderately Impaired

Habitat Analysis: 126

Observations

Streamwater: Slightly Turbid....Flow: Slow....Width/Depth (ft): 25/4

Substrate: Cobbles, gravel, silt.... StreamBank Vegetation/Stability: Trees/Stable

Canopy: Mostly Open....Other: Forested/Rural; Water temp.11.6 /pH 8.1 /DO 7.5 /Cond.522

Upper Delaware Basin - Stockton USGS Quadrangle

Station: AN0093

Plum Bk, Pine Hill Rd, Delaware Twp, Hunterdon County

Date Sampled: 07/16/97

Family	Family Tolerance Value (FTV)	Number of Individuals	
	value (FTV)		
Chironomidae	6	66	
BloodRed Chironomidae	8	16	
Leptophlebiidae	2	5	
Heptageniidae	4	5	
Tubificidae	10	2	
Sialidae	4	2	
Elmidae	4	1	
Gomphidae	1	1	
Physidae	7	1	
Psephenidae	4	1	

Statistical Analysis

Number of Taxa: 10

Total Number of Individuals: 100

% Contribution of Dominant Family: 66.00 % (Chironomidae)

Family Biotic Index: 5.98

Scraper/Filterer Collector Ratio: 0.35

Shredder/Total Ratio: 0.00

E+P+T (Ephemeroptera, Plecoptera, Trichoptera): 2

% EPT: 10.00 EPT/C: 0.12 NJIS Rating: 9

Biological Condition: Moderately Impaired

Habitat Analysis: 170

Observations

Streamwater: Clear....Flow: Slow....Width/Depth (ft): 50/1

Substrate: Cobbles....StreamBank Vegetation/Stability: Trees/Stable

Canopy: Mostly Closed....Other: Forested/Rural; Water temp.16.7 /pH 7.0 /DO 7.3 /Cond.157

Bureau of Freshwater and Biological Monitoring

APPENDIX B STATISTICAL DATA

Sediment Toxicity Tests Watershed Management Areas # 1 and 11 Upper Delaware River Basin

Sediment Toxicity Tests Watershed Management Area # 1

SURVIVAL RESULTS

CONTROL VS. AN0028 (reference)

Survival Proportions with Arc Sine Square Root Transformation

Blank	AN0028	Blank Trans	AN0028 Trans
1.0	0.9	1.4127	1.249
1.0	0.9	1.4127	1.249
1.0	1.0	1.4127	1.4127
1.0	1.0	1.4127	1.4127
1.0	1.0	1.4127	1.4127

Blank Trans	AN0028 Trans	Pooled	Mean	Centered	Ordered	D-value	W-value	Critical-W (0.05)	Result
1.4127	1.249	1.4127		0.0327	-0.131				
1.4127	1.249	1.4127		0.0327	-0.131				
1.4127	1.4127	1.4127	1.38	0.0327	0.0327	0.0429	0.5093	0.842	Not Normal
1.4127	1.4127	1.4127		0.0327	0.0327				
1.4127	1.4127	1.4127		0.0327	0.0327				
		1.249		-0.131	0.0327				
Mean	Mean	1.249		-0.131	0.0327				
1.4127	1.3472	1.4127		0.0327	0.0327				
		1.4127		0.0327	0.0327				
		1.4127		0.0327	0.0327				

Sediment Toxicity Tests Watershed Management Area # 1

SURVIVAL RESULTS continued...

CONTROL VS. AN0028 (reference)

Pooled	Sorted	Point	Wilcoxan Rank	Blank	AN0028	Critical(from Table K=1)	Result
1.4127	1.249	7	1.5	0	1.5	19	No Significant Difference
1.4127	1.249	6	1.5	0	1.5		
1.4127	1.4127	10	6.5	0	6.5		
1.4127	1.4127	9	6.5	0	6.5		
1.4127	1.4127	8	6.5	0	6.5		
1.249	1.4127	5	6.5	6.5	0		
1.249	1.4127	4	6.5	6.5	0		
1.4127	1.4127	3	6.5	6.5	0		
1.4127	1.4127	2	6.5	6.5	0		
1.4127	1.4127	1	6.5	6.5	0		
				Sum	Sum		
·				32.5	22.5		

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

SURVIVAL RESULTS continued...

AN0028 (reference) VS. AN0016

Survival Proportions with Arc Sine Square Root Transformation

AN0028	AN0016	AN0028 Trans	AN0016 Trans
0.9	1.0	1.249	1.4127
0.9	0.7	1.249	0.9912
1.0	0.9	1.4127	1.249
1.0	1.0	1.4127	1.4127
1.0	0.9	1.4127	1.249

AN0028 Trans	AN0016 Trans	Pooled	Mean	Centered	Ordered	D-value	W-value	Critical-W (0.05)	Result
1.249	1.4127	1.249		-0.0561	-0.3139				
1.249	0.9912	1.249		-0.0561	-0.0561				
1.4127	1.249	1.4127	1.3051	0.1076	-0.0561	0.169	0.7558	0.842	Not Normal
1.4127	1.4127	1.4127		0.1076	-0.0561				
1.4127	1.249	1.4127		0.1076	-0.0561				
		1.4127		0.1076	0.1076				
Mean	Mean	0.9912		-0.3139	0.1076				
1.3472	1.2629	1.249		-0.0561	0.1076				
		1.4127		0.1076	0.1076				
		1.249		-0.0561	0.1076				

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

SURVIVAL RESULTS continued...

AN0028 (reference) VS. AN0016

TTHEOREM 1	tank Sum	LCSt					
Pooled	Sorted	Point	Wilcoxan Rank	AN0028	AN0016	Critical(from Table K=1)	Result
1.249	0.9912	7	1	0	1	19	No Significant Difference
1.249	1.249	10	3.5	0	3.5		
1.4127	1.249	8	3.5	0	3.5		
1.4127	1.249	2	3.5	3.5	0		
1.4127	1.249	1	3.5	3.5	0		
1.4127	1.4127	9	8	0	8		
0.9912	1.4127	6	8	0	8		
1.249	1.4127	5	8	8	0		
1.4127	1.4127	4	8	8	0		
1.249	1.4127	3	8	8	0		
				Sum	Sum		
				31	24		

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

SURVIVAL RESULTS continued...

AN0028 (reference) VS. AN0036

<u>Survival Proportions with Arc Sine Square Root Transformation</u>

AN0028	AN0036	AN0028 Trans	AN0036 Trans
0.9	0.9	1.249	1.249
0.9	1.0	1.249	1.4127
1.0	1.0	1.4127	1.4127
1.0	1.0	1.4127	1.4127
1.0	1.0	1.4127	1.4127

	1	1 1 (OI III aii t	/				1	r	
AN0028 Trans	AN0036 Trans	Pooled	Mean	Centered	Ordered	D-value	W-value	Critical-W (0.05)	Result
1.249	1.249	1.249		-0.1146	-0.1146				
1.249	1.4127	1.249		-0.1146	-0.1146				
1.4127	1.4127	1.4127	1.3636	0.0491	-0.1146	0.0563	0.594	0.842	Not Normal
1.4127	1.4127	1.4127		0.0491	0.0491				
1.4127	1.4127	1.4127		0.0491	0.0491				
		1.249		-0.1146	0.0491				
Mean	Mean	1.4127		0.0491	0.0491				
1.3472	1.38	1.4127		0.0491	0.0491				
		1.4127		0.0491	0.0491				
		1.4127		0.0491	0.0491				

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

SURVIVAL RESULTS continued...

AN0028 (reference) VS. AN0036

Pooled	Sorted	Point	Wilcoxan Rank	AN0028	AN0036	Critical(from Table K=1)	Result
1.249	1.249	6	2	0	2	19	No Significant Difference
1.249	1.249	2	2	2	0		
1.4127	1.249	1	2	2	0		
1.4127	1.4127	10	7	0	7		
1.4127	1.4127	9	7	0	7		
1.249	1.4127	8	7	0	7		
1.4127	1.4127	7	7	0	7		
1.4127	1.4127	5	7	7	0		
1.4127	1.4127	4	7	7	0		
1.4127	1.4127	3	7	7	0		
				Sum	Sum		
				25	30		

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

SURVIVAL RESULTS continued...

AN0028 (reference) VS. AN0093

Survival Proportions with Arc Sine Square Root Transformation

AN0028	AN0093	AN0028 Trans	AN0093 Trans
0.9	1.0	1.249	1.4127
0.9	0.8	1.249	1.1071
1.0	0.8	1.4127	1.1071
1.0	0.9	1.4127	1.249
1.0	0.9	1.4127	1.249

AN0028 Trans	AN0093 Trans	Pooled	Mean	Centered	Ordered	D-value	W-value	Critical-W (0.05)	Result
1.249	1.4127	1.249		-0.0371	-0.179				
1.249	1.1071	1.249		-0.0371	-0.179				
1.4127	1.1071	1.4127	1.2861	0.1266	-0.0371	0.1337	0.8197	0.842	Not Normal
1.4127	1.249	1.4127		0.1266	-0.0371				
1.4127	1.249	1.4127		0.1266	-0.0371				
		1.4127		0.1266	-0.0371				
Mean	Mean	1.1071		-0.179	0.1266				
1.3472	1.225	1.1071		-0.179	0.1266				
		1.249		-0.0371	0.1266				
		1.249		-0.0371	0.1266				

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

SURVIVAL RESULTS continued...

AN0028 (reference) VS. AN0093

Pooled	Sorted	Point	Wilcoxan Rank	AN0028	AN0093	Critical(from Table K=1)	Result
1.249	1.1071	8	1.5	0	1.5	19	No Significant Difference
1.249	1.1071	7	1.5	0	1.5		
1.4127	1.249	10	4.5	0	4.5		
1.4127	1.249	9	4.5	0	4.5		
1.4127	1.249	2	4.5	4.5	0		
1.4127	1.249	1	4.5	4.5	0		
1.1071	1.4127	6	8.5	0	8.5		
1.1071	1.4127	5	8.5	8.5	0		
1.249	1.4127	4	8.5	8.5	0		
1.249	1.4127	3	8.5	8.5	0		
				Sum	Sum		
				34.5	20.5		

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

GROWTH

CONTROL VS. AN0028 (reference)

Average Dry Weights (in mg)

Blank	AN0028
0.098	0.176
0.106	0.176
0.130	0.177
0.109	0.176
0.099	0.171

Blank	AN0028	Pooled	Mean	Centered	Ordered	D-value	W-value	Critical-W (0.05)	Result
0.098	0.176	0.098		-0.0438	-0.0438				
0.106	0.176	0.106		-0.0358	-0.0428				
0.130	0.177	0.130	0.1418	-0.0118	-0.0358	0.0118	0.7729	0.842	Not Normal
0.109	0.176	0.109		-0.0328	-0.0328				
0.099	0.171	0.099		-0.0428	-0.0118				
		0.176		0.0342	0.0292				
Mean	Mean	0.176		0.0342	0.0342				
0.108	0.175	0.177		0.0352	0.0342				
		0.176		0.0342	0.0342				
		0.171		0.0292	0.0352				

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

GROWTH continued...

CONTROL VS. AN0028 (reference)

Pooled	Sorted	Point	Wilcoxan Rank	Blank	AN0028	Critical(from Table K=1)	Result
0.098	0.098	1	1	1	0	19	No Significant Difference
0.106	0.099	5	2	2	0		
0.130	0.106	2	3	3	0		
0.109	0.109	4	4	4	0		
0.099	0.130	3	5	5	0		
0.176	0.171	10	6	0	6		
0.176	0.176	9	8	0	8		
0.177	0.176	7	8	0	8		
0.176	0.176	6	8	0	8		
0.171	0.177	8	10	0	10		
				Sum	Sum		
				15	40		

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

GROWTH continued...

AN0028 (reference) VS. AN0016

Average Dry Weights (in mg)

AN0028	AN0016
0.176	0.171
0.176	0.151
0.177	0.208
0.176	0.120
0.171	0.144

Shapiro-Wilks Test for Normality

AN0028	AN0016	Pooled	Mean	Centered	Ordered	D-value	W-value	Critical-W (0.05)	Result
0.176	0.171	0.176		0.009	-0.047				
0.176	0.151	0.176		0.009	-0.023				
0.177	0.208	0.177	0.167	0.010	-0.016	0.005	0.894	0.842	Normal
0.176	0.120	0.176		0.009	0.004				
0.171	0.144	0.171		0.004	0.004				
		0.171		0.004	0.009				
Mean	Mean	0.151		-0.016	0.009				
0.175	0.159	0.208		0.041	0.009				
		0.120		-0.047	0.010				
		0.144		-0.023	0.041				

F-Test and T-Test

AN0028 Var	AN0016 Var	F-Value	Critical-F (Two-Tailed 0.05)	Varianc es	T-value	Deg. of Freedom	Critical-T (One-Tailed 0.05)	Result
0	0.0011	0	6.3882	0	1.1057	4	2.1318	No Significant Difference

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

GROWTH continued...

AN0028 (reference) VS. AN0036

Average Dry Weights (in mg)

AN0028	AN0036
0.176	0.198
0.176	0.197
0.177	0.200
0.176	0.190
0.171	0.196

AN0028	AN0036	Pooled	Mean	Centered	Ordered	D-value	W-value	Critical-W (0.05)	Result
0.176	0.198	0.176		-0.0097	-0.0147				
0.176	0.197	0.176		-0.0097	-0.0097				
0.177	0.200	0.177	0.1857	-0.0087	-0.0097	0.0012	0.8188	0.842	Not Normal
0.176	0.190	0.176		-0.0097	-0.0097				
0.171	0.196	0.171		-0.0147	-0.0087				
		0.198		0.0123	0.0043				
Mean	Mean	0.197		0.0113	0.0103				
0.175	0.196	0.200		0.0143	0.0113				
		0.190		0.0043	0.0123				
		0.196		0.0103	0.0143				

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

GROWTH continued...

AN0028 (reference) VS. AN0036

w ncoxan i	Kank Sum .	i est					
Pooled	Sorted	Point	Wilcoxan Rank	AN0028	AN0036	Critical(from Table K=1)	Result
0.176	0.171	5	1	1	0	19	No Significant Difference
0.176	0.176	4	3	3	0		
0.177	0.176	2	3	3	0		
0.176	0.176	1	3	3	0		
0.171	0.177	3	5	5	0		
0.198	0.190	9	6	0	6		
0.197	0.196	10	7	0	7		
0.200	0.197	7	8	0	8		
0.190	0.198	6	9	0	9		
0.196	0.200	8	10	0	10		
				Sum	Sum		
				15	40		

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

GROWTH continued...

AN0028 (reference) VS. AN0093

Average Dry Weights (in mg)

	J
AN0028	AN0093
0.176	0.144
0.176	0.251
0.177	0.186
0.176	0.180
0.171	0.202

AN0028	AN0093	Pooled	Mean	Centered	Ordered	D-value	W-value	Critical-W (0.05)	Result
0.176	0.144	0.176		-0.0079	-0.0399				
0.176	0.251	0.176		-0.0079	-0.0129				
0.177	0.186	0.177	0.1839	-0.0069	-0.0079	0.0068	0.8114	0.842	Not Normal
0.176	0.180	0.176		-0.0079	-0.0079				
0.171	0.202	0.171		-0.0129	-0.0079				
		0.144		-0.0399	-0.0069				
Mean	Mean	0.251		0.0671	-0.0039				
0.175	0.193	0.186		0.0021	0.0021				
		0.180		-0.0039	0.0181				
		0.202		0.0181	0.0671				

Sediment Toxicity Tests Watershed Management Areas # 1 and 11

GROWTH continued...

AN0028 (reference) VS. AN0093

Pooled	Sorted	Point	Wilcoxan Rank	AN0028	AN0093	Critical(from Table K=1)	Result
0.176	0.144	6	1	0	1	19	No Significant Difference
0.176	0.171	5	2	2	0		
0.177	0.176	4	4	4	0		
0.176	0.176	2	4	4	0		
0.171	0.176	1	4	4	0		
0.144	0.177	3	6	6	0		
0.251	0.180	9	7	0	7		
0.186	0.186	8	8	0	8		
0.180	0.202	10	9	0	9		
0.202	0.251	7	10	0	10		
				Sum	Sum		
				20	35		