BIOASSAYS OF CAPE KENNEDY NASA #1 WASTEWATER TREATMENT PLANT

KENNEDY SPACE CENTER, BREVARD COUNTY, FLORIDA

NPDES #FL0022594

SAMPLED 3/3/81

Biological Section
Bureau of Water Analysis
April 27, 1981

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INTRODUCTION

The Cape Kennedy NASA #1 Wastewater Treatment Plant is located at the Kennedy Space Center in Brevard County, Florida (Fig. 1). The plant has a maximum capacity of 0.37 MGD and serves nearby Air Force and Naval Air Station personnel and their operations. Effluent from this facility flows from the final polishing pond into a storm drainage ditch, then into the Banana River at Outfall #001.

On 4 to 6 March, 1981, the Biological Section performed static toxicity bioassays on effluent samples from this facility to determine the existence and degree of toxicity to the biota of the receiving waters. The results of these tests are presented in this report.

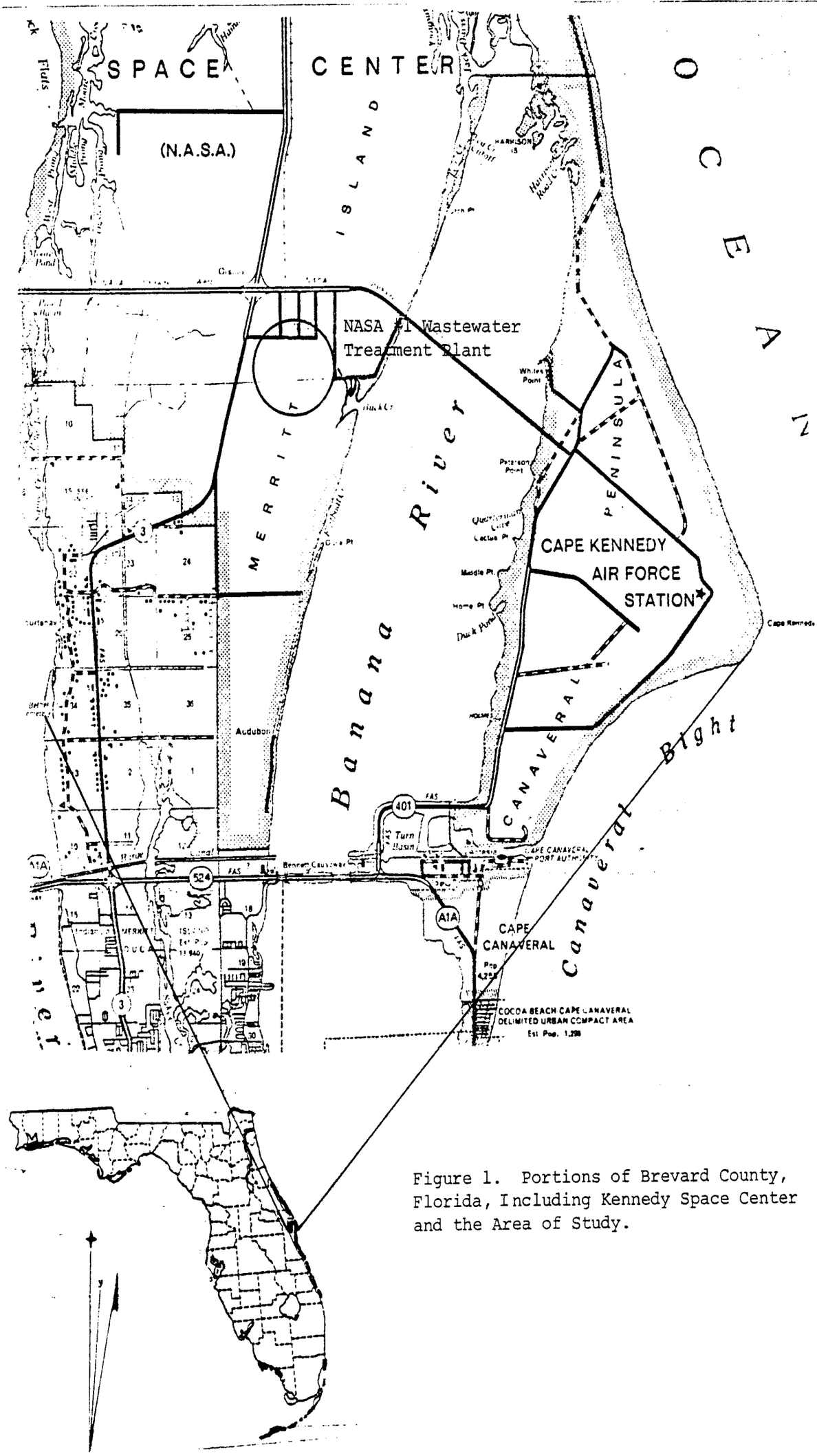


Figure 1. Portions of Brevard County, Florida, Including Kennedy Space Center and the Area of Study.

METHODS AND MATERIALS

Test methods used to conduct these static bioassays are extensively described by Peltier (1978). The organisms selected for this study are among those recommended by Peltier (1978) and the U.S. EPA (1978). Life histories of these test organisms and maintenance procedures used to culture them are detailed below.

History and Maintenance of Bioassay Organisms

Mysidopsis bahia (mysid shrimp) - In September, 1978, approximately 300 adult mysid shrimp were obtained from the Gulf Breeze EPA Laboratory in Pensacola, Florida. The mysids have been cultured under continuous lighting in 10 and 15 gallon all glass aquaria equipped with undergravel filtration and aeration. The mysid shrimp are cultured over a wide range of salinities (10-30 0/00) to minimize acclimation periods to low or high salinity effluents. Sea water is obtained from the FSU Marine Laboratory located at Turkey Point, Florida (salinity range 17-31 0/00) and partial water changes are performed weekly. Fresh water, for dilution of the sea water as necessary, is obtained from a deep well near the DER laboratory. The mysids are fed twice daily with freshly hatched Artemia (brine shrimp) and are kept at a constant temperature of $22^{\circ} \pm 1^{\circ}$C. Immediately prior to the bioassay, a suitable number of mysids were transferred by net to a culture dish for observation and selection of individuals for the test. Fifteen day old (three-quarter adult size) mysids were used in this test. Organisms selected for testing were transferred individually by glass tube and randomly placed in the test chambers.

To ensure the uniform sensitivity of these test organisms to toxic substances, the Biological Section conducts monthly Standard Reference Toxicant Bioassays using sodium lauryl sulfate as the reference toxicant.

Test Methods

Three separate 48 hour static bioassays were conducted from 1030 hours on 4 March, 1981 to 1030 hours on 6 March, 1981. Grab samples of chlorinated effluent (Outfall #001) and of effluent collected prior to chlorination were obtained by Mr. Louis Ley of DER's St. Johns River District Office at 1520 hours on 3 March, 1981, using standard wastewater sampling procedures. These samples were unpreserved. These samples were iced and transported to the DER laboratory in Tallahassee within 24 hours of collection.

To differentiate between the toxicity resulting from chlorine and the toxicity associated with some other component of the effluent, the Biological Section adopted the following test procedure:

> Test #1. Pre-Chlorinated Effluent (Table 2). This test was performed on an effluent sample collected at the end of the treatment process but prior to chlorination. And, was intended to evaluate the toxicity of the effluent without the added toxic effects of chlorine.

Test #2. Chlorinated Effluent (Table 3). This test was performed on an effluent sample collected at the point of discharge.

Test #3. De-Chlorinated Effluent (Table 4). This test was performed on a subsample of the chlorinated effluent sample in which the chlorine had been neutralized[1]. This test was intended to evaluate the combined toxicity of the effluent and the chlorine reaction products.

Saltwater for dilution of the effluent samples was obtained from the FSU Marine Laboratory at Turkey Point (salinity = 31.0 ppt). This saltwater diluent was then adjusted to the salinity of the receiving body of water (Banana River = 25.0 ppt) by adding an appropriate amount of DER well water. For test concentrations of 56% and below, this adjusted diluent was mixed directly with the effluent, achieving a range of final salinities (24 ppt to 12 ppt). For the 100% effluent test concentrations, freshly autoclaved artificial sea salts (Ocean 50®) were added until salinities of approximately 7 ppt were achieved (the lower limit of mysid shrimp salinity tolerances). A series of three control groups (Table 5) were then used to ensure that any mortality in the test organisms was not the result of toxic properties associated with the diluent, the artificial sea salts, or the salinity range. The control groups included: adjusted Turkey Point seawater (salinity = 25.0 ppt), artificial sea salt (Ocean 50®) adjusted DER well water (salinity = 7.0 ppt), and a salinity control consisting of adjusted Turkey Point seawater and DER well water (salinity = 12.5 ppt).

Two replicates of each test concentration and the controls were used in these static tests. Immediately prior to the bioassays, a suitable number of mysids were transferred by net from the 10 ppt, 20 ppt, and 25 ppt salinity aquaria to culture dishes for observation and selection of individuals for the tests. Individual mysid shrimp were siphoned via glass tube, carefully examined for injury, and transferred to wide mouth quart jars, each containing 500ml of the appropriate concentration. For each concentration the mysid shrimp were pre-acclimated to within 4 ppt salinity. The concentration series selected for this study (Table 1) are those recommended by the APHA (1976).

Five organisms were loaded per test chamber, giving ten per concentration. Temperature, pH, dissolved oxygen, and the number of live organisms were recorded at 0, 24, and 48 hours for each test chamber (Tables 2 through 5).

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[1] De-chlorination was accomplished by titrating with an appropriate amount of sodium thiosulfate.

Additionally, ammonia concentrations, including ionized and unionized fractions, were determined for a control and a 100% test concentration at the beginning of the bioassays. According to EPA (1978), "Although it is generally undesirable to feed most fish and macroinvertebrates during static toxicity tests, an exception is necessary for M. bahia". Therefore, the mysids were fed brine shrimp (Artemia) nauplii at the rate of approximately 10 to 20 per mysid shrimp to minimize cannibalism.

Table 1. Concentration Series and Volumes Used for These Static Bioassays

Mysid shrimp (Mysidopsis bahia)

Concentration	Dilution Water	Effluent	Total Vol.
Control	500 ml	---	500 ml
5.6%	472 ml	28 ml	500 ml
10.0%	450 ml	50 ml	500 ml
18.0%	410 ml	90 ml	500 ml
32.0%	340 ml	160 ml	500 ml
56.0%	220 ml	280 ml	500 ml
100%	---	500 ml	500 ml

Table 2 . Data recorded during 48 hour, Mysidopsis bahia, static acute toxicity bioassay of Cape Kennedy NASA #1 Wastewater Treatment Plant, Brevard County, Florida, NPDES #FL0022594, on 4 to 6 March, 1981.

Test #1. Pre-Chlorinated Sample.

Concentration or %	Number of Live Organisms			Dissolved Oxygen (mg/ℓ)			pH			Temperature (°C)			NH_3/NH_3+NH_4 (mg/ℓ)	Salinity (o/oo)
	0	24	48	0	24	48	0	24	48	0	24	48	0	0
5.6%	5[a]	5	5	7.6	6.6	6.8	8.0	7.6	7.3	20.7	19.9	19.6		24.0
5.6%	5[a]	5	5	7.6	6.2	6.7	8.0	7.6	7.3					
10.0%	5[a]	5	5	7.8	7.0	7.0	8.0	7.7	7.3	20.4	19.8	19.5		23.0
10.0%	5[a]	5	5	7.9	6.9	7.0	8.0	7.7	7.3					
18.0%	5[b]	5	5	8.2	7.1	6.9	8.1	7.8	7.4	20.3	19.7	19.5		21.0
18.0%	5[b]	5	5	8.2	7.1	7.0	8.1	7.8	7.4					
32.0%	5[b]	5	5	8.9	7.4	7.1	8.2	7.8	7.4	20.5	19.6	19.5		18.0
32.0%	5[b]	4	4	8.9	7.4	7.2	8.2	7.8	7.4					
56.0%	5[c]	5	5	10.1	8.3	8.1	8.3	8.0	7.6	20.8	19.7	19.5		12.0
56.0%	5[c]	5	5	10.2	8.3	8.1	8.3	8.0	7.6					
100%	6[c,d]	6	6	9.9	8.8	9.6	8.9	8.4	8.0	21.4	19.9	19.7	2.74/10.74[e]	7.0
100%	5[c]	5	5	9.6	9.1	9.7	8.9	8.4	8.0					

Remarks: [a]Mysids cultured at 25 ppt salinity. [c]Mysids cultured at 10 ppt salinity.
[b]Mysids cultured at 20 ppt salinity. [d]To prevent disturbing mysids, the extra organism was allowed to remain.
[e] Total ammonia is expressed as mg/l as N and unionized ammonia is expressed as mg/l as NH_3.

Table 3. Data recorded during 48 hour, Mysidopsis bahia, static acute toxicity bioassay of Cape Kennedy NASA #1 Wastewater Treatment Plant, Brevard County, Florida, NPDES #FL0022594, Outfall #001, on 4 to 6 March, 1981.

Test #2. Chlorinated Sample.

Concentration or %	Number of Live Organisms			Dissolved Oxygen (mg/ℓ)			pH			Temperature (°C)			NH_3/NH_3+NH_4 (mg/ℓ)	Salinity (o/oo)
	0	24	48	0	24	48[d]	0	24	48[d]	0	24	48[d]	0	0
5.6%	5[a]	0	0	8.0	7.9	---	8.0	7.8	---	20.7	19.8	---		23.8
5.6%	5[a]	0	0	8.0	7.9	---	8.0	7.8	---					
10.0%	5[a]	0	0	8.1	8.0	---	8.0	7.8	---	20.3	19.7	---		23.0
10.0%	5[a]	0	0	8.1	8.0	---	8.0	7.8	---					
18.0%	5[b]	0	0	8.4	8.2	---	8.0	7.8	---	20.2	19.6	---		21.0
18.0%	5[b]	0	0	8.5	8.2	---	8.0	7.8	---					
32.0%	5[b]	0	0	8.9	8.4	---	7.9	7.8	---	20.2	19.7	---		18.8
32.0%	5[b]	0	0	9.0	8.4	---	7.9	7.8	---					
56.0%	5[c]	0	0	10.0	8.8	---	7.8	7.7	---	20.5	19.7	---		12.0
56.0%	5[c]	0	0	10.0	8.8	---	7.8	7.7	---					
100%	5[c]	0	0	10.0	9.1	---	8.5	8.0	---	21.1	19.8	---	1.14/9.66[e]	7.0
100%	5[c]	0	0	10.0	9.0	---	8.5	8.0	---					

Remarks: [a]Mysids cultured at 25 ppt salinity.

[b]Mysids cultured at 20 ppt salinity.

[c]Mysids cultured at 10 ppt salinity.

[d]48 hour parameters not taken since all organisms died by 24 hours.

[e]Total ammonia is expressed as mg/l N and unionized ammonia is expressed as mg/l NH_3.

Total chlorine in: 5.6% test=0.4 mg/l (Hach DPD)
10% test = 0.8 mg/l (Hach DPD)
100% test=2.6 mg/l (Hach DPD)

Table 4 . Data recorded during 48 hour, Mysidopsis bahia, static acute toxicity bioassay of Cape Kennedy NASA #1 Wastewater Treatment Plant, Brevard County, Florida, NPDES #FL0022594, Outfall #001, on 4 to 6 March, 1981.

Test #3. De-Chlorinated Sample.

Concentration or %	Number of Live Organisms			Dissolved Oxygen (mg/ℓ)			pH			Temperature (°C)			NH_3/NH_3+NH_4 (mg/ℓ)	Salinity (o/oo)
	0	24	48	0	24	48	0	24	48	0	24	48	0	0
5.6%	5[a]	5	5	8.1	6.0	7.0	8.1	7.7	7.3	20.4	19.8	19.7		23.8
5.6%	5[a]	5	5	8.0	6.1	6.9	8.1	7.7	7.3					
10.0%	5[a]	5	5	8.1	6.1	6.8	8.0	7.7	7.3	20.5	19.8	19.7		23.0
10.0%	5[a]	5	5	8.1	6.1	6.8	8.0	7.7	7.3					
18.0%	5[b]	5	5	8.2	6.0	6.7	8.0	7.7	7.3	20.2	19.8	19.8		21.0
18.0%	5[b]	4(5[d])	4(5[d])	8.3	6.1	6.7	8.0	7.7	7.3					
32.0%	5[b]	5	5	8.7	5.7	6.6	8.1	7.6	7.3	19.9	19.9	20.0		17.5
32.0%	5[b]	5	5	8.8	5.7	6.6	8.1	7.6	7.3					
56.0%	5[c]	5	5	9.5	5.5	6.8	8.1	7.6	7.2	20.2	19.9	19.8		12.0
56.0%	5[c]	5	5	9.5	5.4	6.9	8.1	7.6	7.2					
100%	5[c]	5	5	10.1	6.1	6.6	8.7	8.2	7.5	20.8	20.1	20.1		7.0
100%	5[c]	5	5	10.1	6.1	6.6	8.8	8.2	7.5					

Remarks:

[a] Mysids cultured at 25 ppt salinity.

[b] Mysids cultured at 20 ppt salinity.

[c] Mysids cultured at 10 ppt salinity.

[d] Mortality due to cannibalism, therefore, number in parentheses used in calculations.

Table 5 . Data recorded during 48 hour, Mysidopsis bahia, static acute toxicity bioassay of Cape Kennedy NASA #1 Wastewater Treatment Plant, Brevard County, Florida, NPDES #FL0022594, on 4 to 6 March, 1981.

Test #4. Control Series.

Concentration or %	Number of Live Organisms			Dissolved Oxygen (mg/ℓ)			pH			Temperature (°C)			NH_3/NH_3+NH_4 (mg/ℓ)	Salinity (o/oo)
	0	24	48	0	24	48	0	24	48	0	24	48	0	0
Seawater Control	5[a]	5	5	7.7	7.0	5.9	7.9	7.6	7.1	20.4	20.0	19.9		25.0
Seawater Control	5[a]	5	5	7.7	7.0	5.9	7.9	7.6	7.1					
Seasalt Control	5[b]	5	5	8.4	8.2	7.4	8.6	8.3	7.6	22.0	20.0	19.8		7.0
Seasalt Control	5[b]	5	5	8.4	8.5	7.4	8.6	8.3	7.6					
Salinity Control	5[b]	5	5	5.3	7.2	7.5	8.1	7.4	7.0	22.0	20.0	20.1	.03/.18[c]	12.5
Salinity Control	5[b]	5	5	5.3	7.2	7.4	8.1	7.4	7.0					

Remarks:

[a] Mysids cultured at 25 ppt salinity.

[b] Mysids cultured at 10 ppt salinity.

[c] Total ammonia is expressed as mg/l as N and unionized ammonia is expressed as mg/l as NH_3.

RESULTS AND DISCUSSION

Neither the pre-chlorinated (Table 2) nor the de-chlorinated (Table 4) samples from the Cape Kennedy NASA #1 Wastewater Treatment Plant exhibited toxicity to the bioassay test organism, Mysidopsis bahia. However, the chlorinated sample (Table 3) exhibited extreme toxicity to the mysid shrimp. In this test all of the shrimp died within 24 hours at every concentration. The 24, 48 and 96 hour LC_{50}'s for this bioassay would therefore be <5.6%. This toxicity can be directly attributed to the presence of chlorine since a residualof 2.6 mg/l was found in the 100% effluent, substantially above the tolerance limit of Mysidopsis bahia.

Chapter 17-3.051, Florida Administrative Code, indicates that "all waters of the State shall at all places and at all times be free from...components of discharges which...are acutely toxic." "Acute toxicity" is defined in Chapter 17-3.021(1) to mean "the presence of one or more substances...in amounts which are greater than one-third (1/3) of the amount lethal to 50% of the test organisms in 96 hours (96 hour LC_{50})...". In the absence of a mixing zone, this 1/3 of the 96 hour LC_{50} applies at the point of discharge. Chapter 17-4.244(4) states that "in no event shall the maximum concentration of wastes in the mixing zone exceed the amount lethal to 50% of the test organisms in 96 hours (96 hour LC_{50}) for a species significant to the indigenous aquatic ommunity..." This criterion applies at the point of discharge if there is a mixing zone. The distinction between presence or absence of a mixing zone is not relevant here, however, since the testing procedure used does not involve effluent concentration. The 1/3 of the 96 hour LC_{50} criterion is three times as stringent as the 96 hour LC_{50} criterion, and therefore, in the absence of a mixing zone, the procedure used is very lenient. The procedure followed is also lenient in that testing is only done for 48 hours. The 96 hour LC_{50} derived from this this testing is therefore reported as "less than or equal to" the 48 hour measurement. The value found after 48 hours can never be more restrictive than the value found after 96 hours. It must be noted that all organisms used in these tests are significant to the indigenous aquatic community since they are sensitive to toxic substances which may be expected to impact the indigenous community.

The toxicity reported here is in excess of allowable limits.

LITERATURE CITED

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