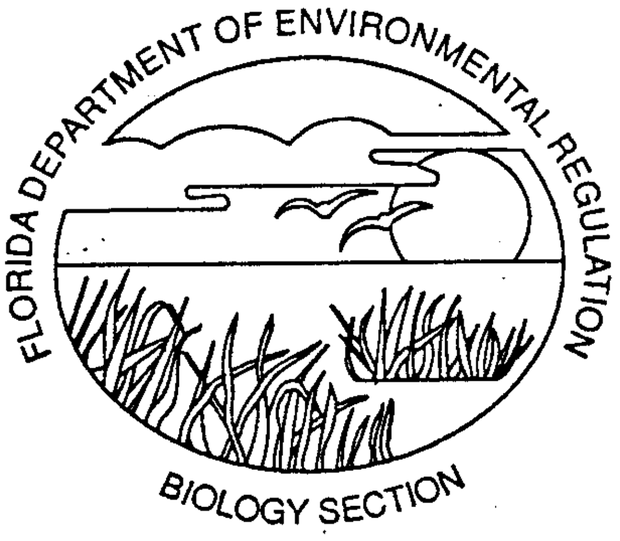


Bioassays of

Gardinier Incorporated

Tampa, Hillsborough County, Florida

NPDES #FL0000761

Sampled 8/8/88

OUTFALL 005

February 16, 1989

Biology Section
Division of Administrative and Technical Services

Bioassays of

Gardinier Incorporated

Tampa, Hillsborough County, Florida

NPDES #FL0000761

Biology Section
Bureau of Laboratories
February 16, 1989

Executive Summary

Gardinier Incorporated, Tampa, Hillsborough County, Florida, NPDES #FL0000761 on 9 to 11 August, 1988.

The sample of discharge collected from outfall #005 of the Gardinier facility was not toxic to the bioassay test organisms, *Ceriodaphnia dubia*, *Daphnia pulex* and *Notropis leedsi*, during the first twenty-four hour test period. At the end of the second twenty-four hour test period, deaths were noted in each of the 100% sample tests. The tests were all invalidated at 48 hrs, however, since dissolved oxygen levels fell below the minimum allowable 40% of saturation. The deaths of *N. leedsi* were thought to have been the result of the low concentrations of dissolved oxygen induced by biochemical oxygen demand. Low dissolved oxygen levels were not thought to be the cause of the deaths noted in the *C. dubia* test. The determination of toxicity in the sample was inconclusive. The sample was stimulatory to the Microtox® assay test organism, *Photobacterium phosphoreum*.

Chemical analyses of the sample detected the metals; arsenic (10 µg/l), chromium (49 µg/l), copper (32 µg/l), silver (0.26 µg/l), and zinc (172 µg/l). The concentrations of these metals were not thought to have contributed to the observed deaths at 48 hours.

The determination of toxicity in this sample was inconclusive due to a biochemical oxygen demand which caused dissolved oxygen levels to decrease below the point of test validity

Introduction

Gardinier, Inc., is located at U.S. 41 and Riverview Drive, Tampa, Hillsborough County, Florida, just north of the mouth of the Alafia River on Hillsborough Bay, (Figure 1). This facility manufactures fertilizer and inorganic chemicals, including sulfuric, phosphoric, and fluorosilic acids, triple super and super phosphate, ammonium phosphate, anhydrous ammonia, and gypsum. Wastewater from the various operations is treated by liming and retention in sludge ponds. As many as six NPDES permitted outfalls are located on the site, three (#001, #005, and #012) that discharge to surface waters. Outfalls #001 and #005 contain water from the sulfuric acid plant and discharge to the Alafia River. Outfall #012 contains supernatant from the sludge ponds and discharges to Archer Creek, a tributary of Hillsborough Bay. A schematic of the facility and its wastewater outfalls is shown in Figure 2.

In 1973, Gardinier, Inc. was fined for damaging public lands (DER NPDES file). Since then, major modifications were introduced in the manufacturing of phosphate chemicals, including a new gypsum disposal field (Figure 3). These modifications reduced energy consumption and increased efficiency and production.

In September 1981, the Biology Section performed bioassays on samples from Outfalls #001, #005, and #012. Some mortality occurred in the test organism, *Mysidopsis bahia*, but an LC_{50} could not be calculated. Chemical analysis indicated that zinc and selenium were presented in the sample from Outfall #005 (FDER 1987).

In June, 1987, a bioassay was performed on a sample from Outfall #005 of this facility. This sample produced an LC_{50} of 376.6% in the test organism, *Mysidopsis bahia.* Metals and unionized ammonia were thought to have contributed to the observed mortality in the test organism (FDER 1987).

In March, 1988, three acute toxicity bioassays and one Microtox® assay were performed on a sample from Outfall #005 of the Gardinier facility. The sample was acutely toxic to the test organism, *Ceriodaphnia dubia*, producing an LC_{50} of 84.9%. The sample was not toxic to the bioassay test organisms, *Daphnia pulex* and *Notropis leedsi*, or to the Microtox® assay organism, *Photobacterium phosphoreum.* The metal, zinc, in a concentration of 439 µg/l, was thought to have contributed to the deaths in the *C. dubia* tests (FDER 1988).

To again evaluate the existence and extent of toxicity in the discharge from Outfall #005 of this facility, the Biology Section performed three screening acute toxicity bioassays and one Microtox® assay on a sample during August, 1988. The results of these tests are presented in this report. Those involved in performing the tests and in preparation of this report were: J. M. Faircloth, R. B. Frydenborg, K. M. Lurding, L. A. Williams, and S. H. Wolfe.

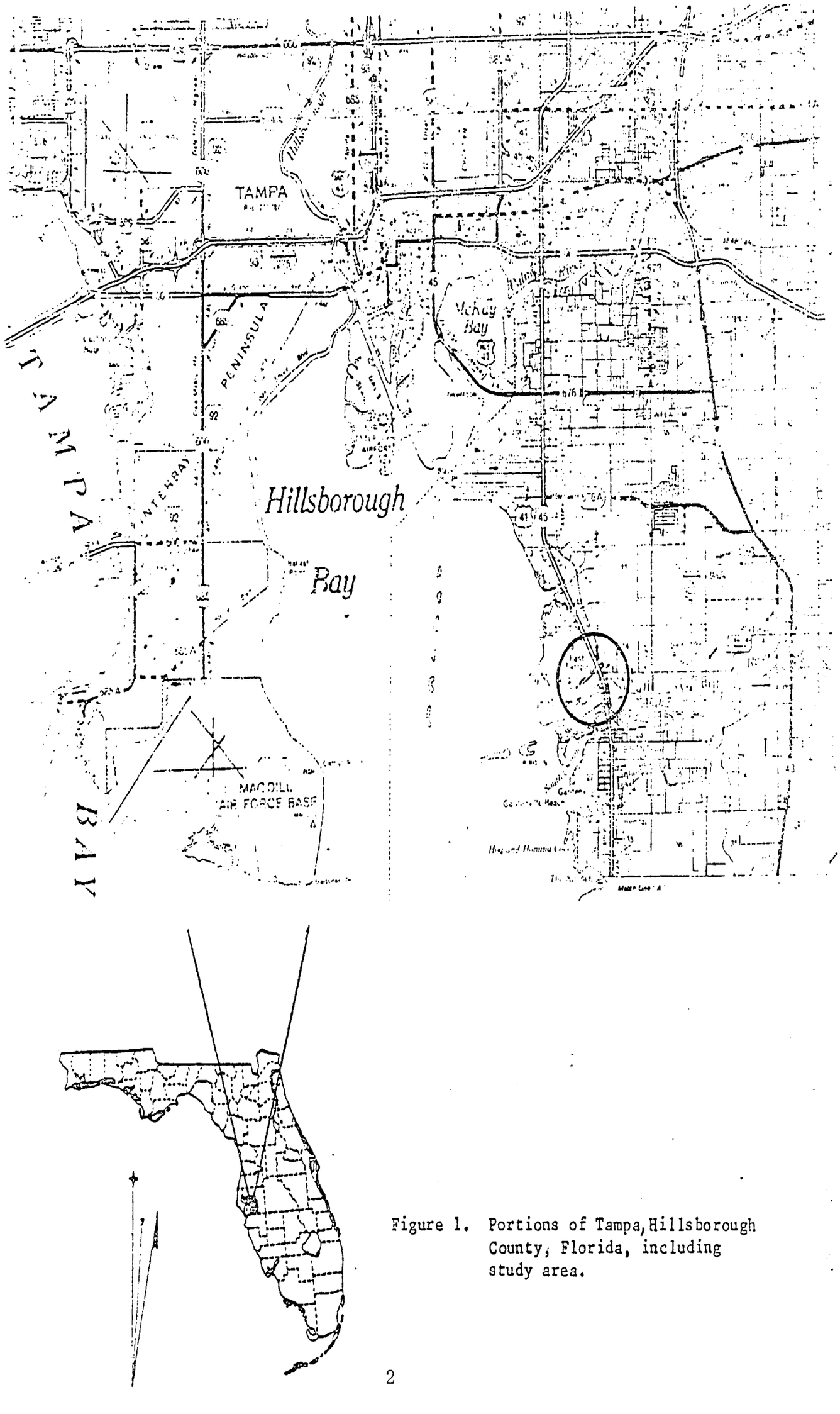
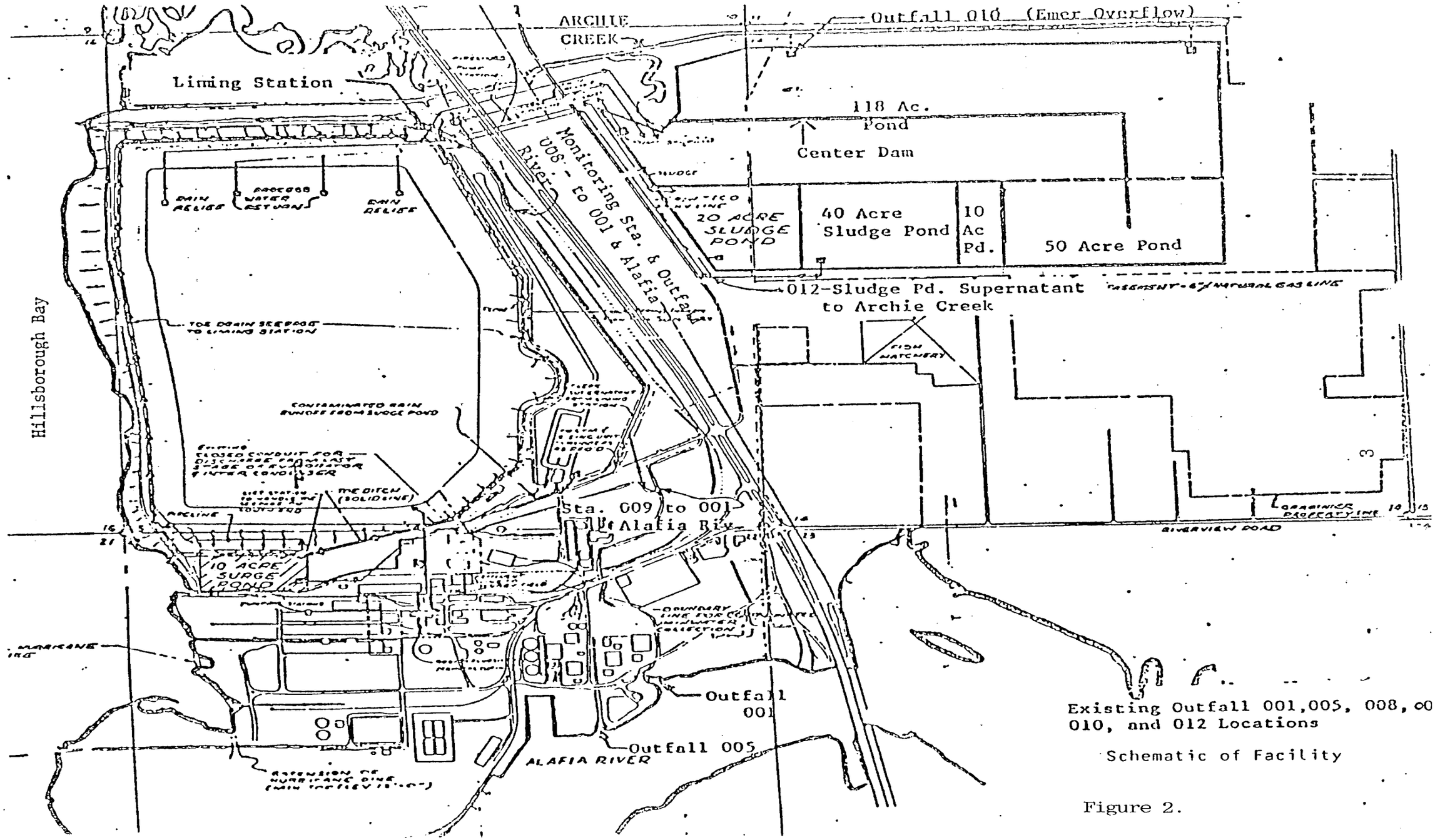


Figure 1. Portions of Tampa, Hillsborough County, Florida, including study area.



Existing Outfall 001,005, 008, 00
010, and 012 Locations

Schematic of Facility

Figure 2.

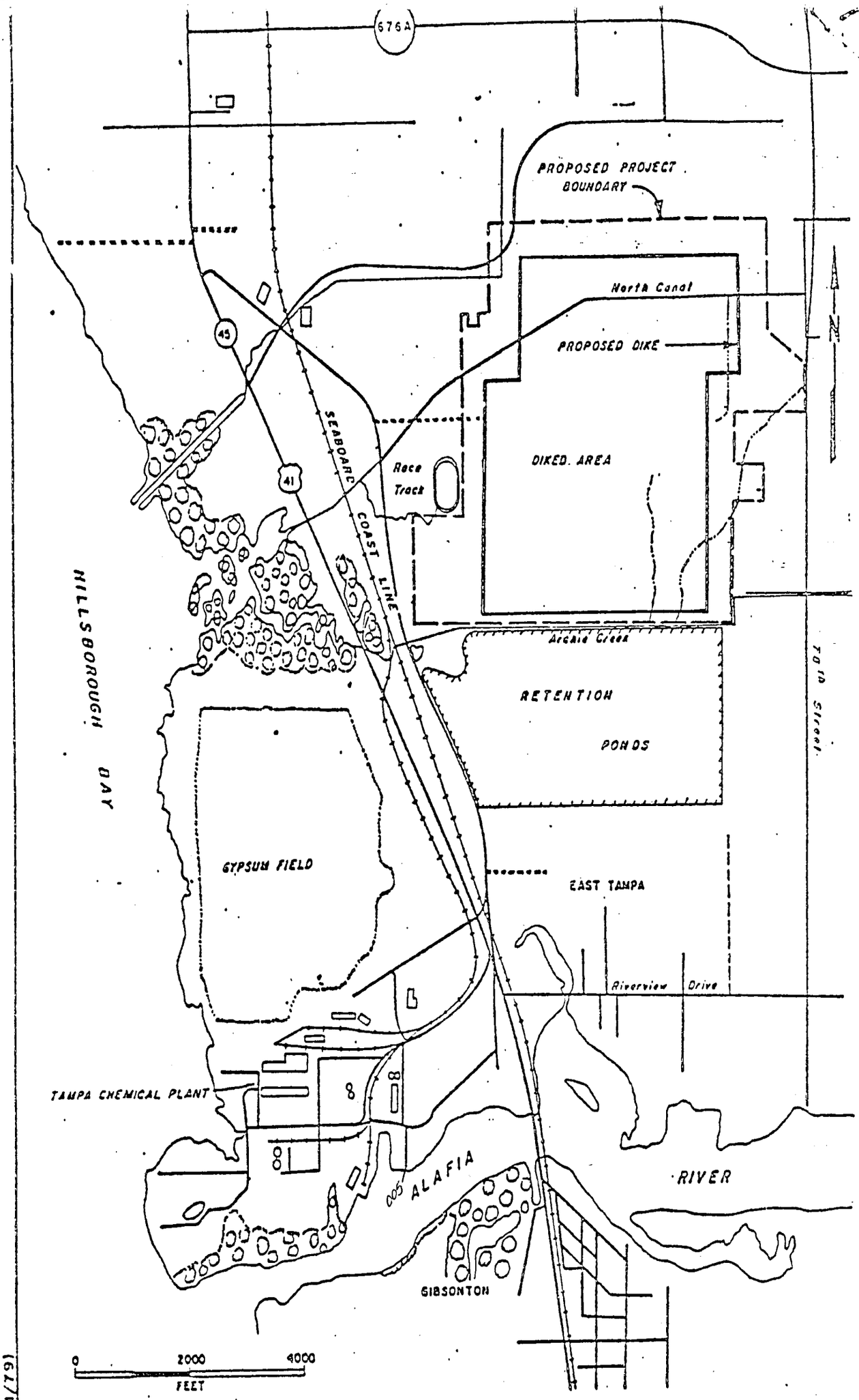


Figure 3 GYPSUM DISPOSAL FIELD LOCATION MAP

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Methods and Materials

Test methods used to conduct these static bioassays are extensively described by Peltier and Weber (1985) and the FDER Biology Section (1985). The organisms selected for this study include a species recommended by Peltier and Weber (1985) and the FDER Biology Section (1985). Additionally, the Biology Section maintains cultures of several species of organisms indigenous to the State of Florida. One of these indigenous species was also selected for testing. Life histories of these test organisms and maintenance procedures used to culture them are detailed below.

The Microtox® assay was run according to the manufacturer's protocol described in the Microtox® System Operating Manual (Beckman 1982).

1. History and Maintenance of Bioassay Organisms

Daphnia pulex (water flea)—A breeding culture of *Daphnia pulex* was obtained from the EPA laboratory in Athens, Georgia, in June, 1987. This culture has been maintained since its arrival in 2 1/2 gallon all-glass aquaria, filled with unchlorinated DER well water, under a 16:8 hour light-dark cycle. Organisms are fed a liquified blend of Purina Trout Chow® and Cerophyl® twice daily, once daily on weekends, and are kept at a constant temperature of 22 ± 2 °C. The aquaria are cleaned weekly by siphoning off half the water along with the detritus and excess organisms, and then refilling with fresh aerated well water. Additional measures are taken and/or new cultures started in the event of abnormal circumstances such as: declining populations, ephippia formation, or excessive fungal or bacterial growth.

Ceriodaphnia dubia (water flea)—A breeding culture of *Ceriodaphnia dubia* was obtained from the EPA laboratory in Athens, Georgia, in June, 1985. The culture is maintained in 2-1/2 gallon all-glass aquaria, filled with unchlorinated DER well water, under a 16:8 hour light-dark cycle. Organisms are fed a liquified blend of Purina Trout Chow®, Cerophyl® and Fleischmann's® yeast twice daily, once daily on weekends, and are kept at a constant temperature of 26 ± 2 °C. The aquaria are cleaned weekly by siphoning off half the water along with the detritus and excess organisms, and then refilling with fresh aerated well water. Additional measures are taken and/or new cultures started in the event of abnormal circumstances such as: declining populations, ephippia formation, or excessive fungal or bacterial growth.

Notropis leedsi (bannerfin shiner)—Approximately 40 adult bannerfin shiners were obtained from an indigenous population in the upper Suwannee River during October, 1981. The breeding stock is maintained in 125 gallon all-glass aquaria equipped with a flow-through water exchange system. Water for this system is unchlorinated and is pumped from the DER deep well. The bannerfin shiners are reared at 22 ± 2 °C under a 16:8 hour light-dark cycle and are fed Aquarian® flake food and frozen *Artemia* (brine shrimp) twice daily, and once daily on weekends. Breeding and egg hatching are encouraged at 25 °C. The eggs are laid between stacked red ceramic tiles separated by a 3mm bead of silicone. Most of the eggs adhere to the tiles and are easily removed to heated, aerated hatching tanks. The narrow space between the tiles eliminates egg predation by adult fish. New hatching tanks are started regularly to segregate fry into even age groups. Newly hatched fry are fed a liquefied blend of Purina Trout Chow® and Cerophyl® twice daily. After one week, feeding is augmented with newly hatched *Artemia* nauplii and finely crushed Aquarian® flake food. The fish used were 17 days old when the test commenced.

Photobacterium phosphoreum, a luminescent marine bacterium, is maintained as a freeze-dried culture, purchased from Microbics Corporation. Lot number M-606 was used in this assay.

2. Standard Reference Toxicity Assays

To ensure the uniform sensitivity of these test organisms to toxic substances, the Biology Section conducts monthly standard reference toxicant bioassays using sodium lauryl sulfate as the reference toxicant.

3. Test Methods

Methods used to perform acute toxicity bioassays are described by Peltier and Weber (1985) and the FDER Biology Section (1985). Some of the methods described below are more specific than those of Peltier and Weber. These will be noted in more detail.

Sampling Methods and Test Dates

Three separate 48 hour acute toxicity screening bioassays and one Microtox® assay were performed on a sample from outfall #005 of the Gardinier facility. The tests were performed from 1100 hours on 9 August, 1988 to 1100 hours on 11 August, 1988.

Separate samples were obtained for chemical and biological testing. Samples for metal analyses were preserved with HNO_3. Samples for organic chemical analyses and for the bioassays were unpreserved. The samples were collected by Mr. David Demmi of the FDER SW District Office in Tampa at 1041 hours on 8 August, 1988, using standard wastewater sampling procedures. The samples were iced and transported to the DER laboratory in Tallahassee for analyses within 24 hours of collection.

Test Temperature

Upon arrival of the iced (4 °C) bioassay samples in the Tallahassee laboratory, the containers were placed in warm water and the temperature of the samples allowed to rise to 20 ± 2 °C. Although this temperature may be different than the temperature of the sample during collection, it is the temperature range to which the test organisms are acclimatized and is the range recommended in the above referenced bioassay protocols. This avoids stressful effects caused by exposure to unusually cold or warm samples.

Method

Freshwater for diluting the water samples being tested was obtained from the same DER deep well that is used to culture the test organisms. The concentration series selected for this study is given in Table 1.

Four replicates of the control and each test concentration were used in bioassays with the water flea, *Ceriodaphnia dubia*. Two replicates of the control and each test concentration were used in bioassays with the water flea *Daphnia pulex*. Plastic vials of 30 ml (for *C. dubia*) capacity and flint glass jars of 250 ml (for *D. pulex*) capacity were used as test chambers, and either 15 ml or 100 ml, respectively, of the appropriate test concentration was placed in each chamber. *Ceriodaphnia* were siphoned into a large culture dish and neonates were transferred by glass pipette. Five were loaded per chamber, resulting in 20 per test concentration. *Daphnia* were siphoned into a stainless steel screen chamber placed into a large culture dish to isolate the proper size and age organisms (neonates). Then the screen chamber along with any retained adults were removed. The neonates remaining were then captured by glass pipette, examined for injury, and loaded 10 per chamber, resulting in 20 per test concentration. The water fleas were not fed during these tests.

Two replicates of each concentration and a control were used for the *Notropis leedsi* (bannerfin shiner) bioassay. The fish were transferred individually or by twos using a pipette into wide mouth quart jars containing 500 ml each of the appropriate test concentration. Five were loaded per chamber, resulting in 10 per concentration. Fish were not fed during this test.

Test Parameters

Dissolved oxygen (YSI Model 57 oxygen meter), pH (Corning Model 7 pH meter with Orion Ross® combination electrode), temperature (NBS calibrated glass thermometer), and the number of live organisms were recorded for each test concentration at 0 hours, 24 hours, and 48 hours. Total alkalinity and total hardness (measured as $CaCO_3$ using Hach Chemical Company reagents) were recorded at the beginning of the test for the controls and the 100% reclaimed water concentration. The specific conductance (YSI Model 33 S-C-T meter) of each test concentration was recorded at the beginning and end of the test. All instruments were calibrated before use according to manufacturer's recommendations. Ammonia concentrations, including the total and unionized fraction (expressed in mg/l as N and as NH_3, respectively), were recorded in the controls and the 100% reclaimed water test concentration at the beginning of the test. Total ammonia concentrations were determined using an Orion specific ion probe and an Orion Ionalyzer®. The unionized fraction was then calculated based on the total ammonia concentration and the pH, temperature, and salinity of the sample. These values were recorded on the bioassay data forms and are presented in Table 2.

Microtox® Test

Upon receipt at the DER laboratory, a 20 ml subsample was collected for Microtox® analysis. The sample was adjusted to 2% salinity with reagent grade dry sodium chloride to maintain the osmotic balance of the test bacterium during the assay. Four two-fold dilutions were made of the test sample resulting in a test range of 99%, 49.5%, 24.75%, 12.38%, and 0%. An incubation temperature of 15 °C was maintained throughout the assay. Ten µl of bacterial suspension was added to each of the 1.0 ml test dilutions. Light output (luminescence) was read after 5 and 15 minutes incubation, using the Microtox® analyzer and recorded on a Perkin-Elmer strip chart recorder. In the presence of a toxicant, the luminescence of the test organisms will decrease. In the presence of a stimulant, the luminescence will increase. Toxicity is calculated as a function of the light lost in the test sample, normalized to the control, and is reported as the effective concentration at which a 50% reduction, of light output is determined in 15 minutes (EC_{50}). Results are given in Table 3.

Results and Discussion

The sample of discharge from outfall #005 of Gardinier, Inc., was not toxic to the bioassay test organisms, *Ceriodaphnia dubia*, *Daphnia pulex*, and *Notropis leedsi*, at the end of the first 24 hour test period. The sample was stimulatory to the Microtox® assay organism, *Photobacterium phosphoreum* (Table 3).

During the second 24 hour test period, a biochemical oxygen demand developed which lowered the dissolved oxygen concentrations in the test chambers (100% sample) below the allowable minimum (Table 2). The deaths which were recorded in *N. leedsi* bioassay were presumed to have been caused by the low dissolved oxygen levels. While the deaths observed in the *C. dubia* were probably not caused by low DO, the test invalidation renders the results inconclusive.

Chemical analyses of the sample detected the metals, arsenic, chromium, copper, silver, and zinc (Table 5). These metals at the detected concentrations were not thought to have contributed to the deaths noted in the test organisms. The total ammonia (2.89 mg/l) and unionized ammonia (0.01 mg/l) concentrations were also considered non-contributory to the recorded deaths.

The presence or absence of toxicity was not determined in the sample taken from the Gardinier facility outfall #005 on 8 August, 1988.

Biology Section Recommendations

Due to the inconclusive nature of these tests and the toxicity reported in earlier tests, the Biology Section recommends that this facility continue to be periodically monitored through bioassays and chemical analyses.

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

Ceriodaphnia dubia (water flea)

Concentration	Dilution Water	Sample	Total Volume
Control	15.0 ml	0.0 ml	15 ml
100.0%	0.0 ml	15.0 ml	15 ml

Daphnia pulex (water flea)

Concentration	Dilution Water	Sample	Total Volume
Control	100.0 ml	0.0 ml	100 ml
100.0%	0.0 ml	100.0 ml	100 ml

Notropis leedsi (bannerfin shiner)

Concentration	Dilution Water	Sample	Total Volume
Control	500 ml	0 ml	500 ml
100.0%	0 ml	500 ml	500 ml

Table 2. Data recorded during 48 hour static acute toxicity screening bioassay of a sample from outfall #005 of the Gardinier, Incorporated, facility, Tampa, Hillsborough County, Florida, NPDES #FL0000761, on 9 to 11 August, 1988.

Facility:	**Gardinier Incorporated**		**NPDES #**	**FL0000761**	Facility Type: **Industrial**
Location:	Hwy 41 Tampa		Contact/District:	D. Demmi	
County:	Hillsborough		Test type:	Static Screening	
Sample Collection Date:	**8/8/88**	Time: 1041	# tests:	3	Receiving Water: Alafia River
Test Beginning Date:	8/9/88	Time: 1100	Chlorination Type:	**nonchlorinated**	
Test Ending Date:	8/11/88	Time: 1100			

Total Residual Cl2	mg/l	Method
Field:	0.0	-
Lab:	0.0	Hach
Lab:	-	-

Ammonia	Total (mg/l)	Unionized (mg/l)
Control Water (fish):	0.00	0.00
Cond. Control Water (daphnids):	0.00	0.00
100% Sample:	2.89	0.01

Alk & Hardness	Alkalinity (mg/l)	Hardness (mg/l)
Control Water (fish):	120	122
Cond. Control Water (daphnids):	118	120
100% Sample:	85	580

Salinity (ppt)	
Control Water:	0
100% Sample:	0

Organism: ***Ceriodaphnia dubia*** Life stage: neonate
LC50: - Binomial: - Moving Average: - Probit: -

	SURVIVAL			Dissolved Oxygen			pH			Temperature			Conductivity	
Concentrations	# at 0 hr	Live at 24 hr	Live at 48 hr	0 hour	24 hour	48 hour	0 hour	24 hour	48 hour	0 hour	24 hour	48 hour	0 hour	48 hour
Control	5	5	5	8.7	-	8.6	8.1	-	8.0	19.4	-	18.8	-	-
Control	5	5	5	8.8	-	8.7	8.1	-	8.0	-	-	-	-	-
Control	5	5	5	8.8	-	8.6	8.1	-	8.1	-	-	-	-	-
Control	5	5	5	8.9	-	8.7	8.1	-	8.1	-	-	-	205	-
100%	5	5	4	7.9	-	7.0	6.8	-	7.3	19.0	-	19.0	-	-
100%	5	5	4	7.9	-	4.6	6.8	-	7.1	-	-	-	-	-
100%	5	5	3	7.9	-	2.8	6.8	-	7.1	-	-	-	-	-
100%	5	5	5	7.9	-	2.8	6.8	-	7.1	-	-	-	2850	-

Organism: ***Daphnia pulex*** Life stage: neonate
LC50: - Binomial: - Moving Average: - Probit: -

	SURVIVAL			Dissolved Oxygen			pH			Temperature			Conductivity	
Concentrations	# at 0 hr	Live at 24 hr	Live at 48 hr	0 hour	24 hour	48 hour	0 hour	24 hour	48 hour	0 hour	24 hour	48 hour	0 hour	48 hour
Control	5	10	10	8.9	8.6	8.9	8.2	7.9	8.0	19.9	19.9	19.3	-	-
Control	5	10	10	8.9	8.7	8.9	8.2	8.0	8.1	-	-	-	205	223
100%	5	10	9	7.1	8.8	1.8	6.8	7.6	7.0	19.3	19.3	19.2	-	-
100%	5	10	9	7.1	8.8	1.5	6.8	7.6	7.0	-	-	-	2850	2560

Organism: ***Notropis leedsi*** Age: 0 days
LC50: - Binomial: - Moving Average: - Probit: -

	SURVIVAL			Dissolved Oxygen			pH			Temperature			Conductivity	
Concentrations	# at 0 hr	Live at 24 hr	Live at 48 hr	0 hour	24 hour	48 hour	0 hour	24 hour	48 hour	0 hour	24 hour	48 hour	0 hour	48 hour
Control	5	5	5	8.9	8.0	8.1	8.2	7.8	7.8	20.0	19.3	19.0	-	-
Control	5	5	5	9.0	8.1	8.1	8.2	7.9	7.9	-	-	-	230	228
100%	5	5	1	6.8	7.6	2.9	6.7	7.2	7.1	19.2	19.4	19.1	-	-
100%	5	6	2	6.8	7.6	2.9	6.7	7.2	7.1	-	-	-	3090	3090

Table 3. Microtox test results, Gardinier Incorporated, Hillsborough County, NPDES #FL0000761, on 9 August, 1988.

Microtox Test — Culture Lot # M-606

% Sample	Final Reclaimed Water nonchlorinated 5-min	15-min
0	73	50
0	72	52
0	70	44
12.38	83	58
12.38	80	58
12.38	83	57
24.75	83	71
24.75	90	70
24.75	93	75
49.5	90	71
49.5	92	73
49.5	92	74
99	91	76
99	90	79
99	95	82

EC_{50} Nonchlorinated (15 minute, 15 °C) = stimulatory

EC_{50} (15 minute, 15 °C) is calculated using the Data Reduction Computer Program developed by Microbics Corporation for the Sharp Model EL-5100 (Microbics) Systems Operating Manual, 1982.

Table 5. Chemical components detected in the sample of discharged process water from the Gardinier facility, Tampa, Hillsborough County, Florida, NPDES #FL0000761, on 9 August, 1988.

Organics	
Acid Extractables	None Detected
Base Neutral Extractables	None Detected
Pesticide Extractables	None Detected

Metals	
Arsenic	10 µg/l
Chromium	49 µg/l
Copper	32 µg/l
Silver	0.26 µg/l
Zinc	172 µg/l

Literature Cited

Beckman Instruments, Inc. 1982. Microtox system operating manual.

Florida Department of Environmental Regulation. 1982. Bioassay of Gardinier, Inc., U.S. Phosphoric Products, Tampa, Hillsborough County, Florida, NPDES #FL0000761. FDER. 15 p.

_____. 1985. Quality assurance manual for performing acute toxicity tests (Second Edition). Biology Section, Tallahassee.

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_____. 1988. Bioassay of Gardinier, Inc., U. S. Phosphoric Products, Tampa, Hillsborough County, Florida, NPDES #FL0000761. FDER. 22 p.

Peltier, W. and C. I. Weber. 1985. Methods for measuring the acute toxicity of effluents to aquatic organisms (Third Edition). Environmental Research Laboratory. Athens, Georgia. EPA-600/4-85-012. 217 p.

Gardinier

Hwy 41

005

Test Number: 1 of 3

Test Type: (Definitive) Renewal (Screening) Conventional

Design:

Sample Collected: Date 8-8-88 1041

Most Recently: Date 8-9-88 1100

Test Ended: Date 8-11-88 1100

Test Organism: Ceriodaphnia dubia

Age/Life Stage: neonate

Test Conc. or Control	Test Container Number	Number of Organisms Alive 0	24	48	72	96	Dissolved Oxygen (mg/l) 0	24	48	72	96	pH 0	24	48	72	96	Temperature °C 0	24	48	72	96	Conductivity 0	24	48	72	96
Control	1	5	5	5			8.7		8.6			8.1	8.0	8.0			19.4		18.8							
Control	2	5	5	5			8.8		8.7			8.1		8.0								205				
Control	3	5	5	5			8.8		8.6			8.1		8.1												
Control	4	5	5	5			8.9		8.7			8.1		8.1								1850				
100%	5	5	5	4			7.9		7.0*			6.8		7.3			19.0		19.0							
100%	6	5	5	4			7.9		4.6*			6.8		7.1												
100%	7	5	5	3			7.9		2.8*			6.8		7.1												
100%	8	5	5	5			7.9		2.6*			6.8		7.1												
Initial of Investigator		KW	KMH	MF			SHW		MF			SHW		MF			SHW		MF			SHW				

Alkalinity/Hardness (mg/l as CaCO3)

Control Water: 118 120

100% Sample: 85 580

Other:

Measured by: MF

Ammonia unionized/total (mg/l as NH3/N)

Hour 0

Control Water: 0.00

100% Sample: 2.89

Other:

Hour 24

Control Water:

100% Sample:

Other:

Measured by:

slope = 55.0

Signatures:

KM Lurding

Mary Ann Faucett

Steven H Wolfe

Russell Frydenborg

Instrument Calibrations:

	pH Meter:	DO Meter:	S-C-T Meter:
0 Hours			
24 Hours			
48 Hours			
72 Hours			
96 Hours			

Remarks: * checked 2X.

Total Residual Chlorine:

Field data: _____ Method: _____

Lab: 0.0 Method: HACH

Lab: _____ Method: _____

De-chlorinated:

Measurements by: KML

Reviewed by:

Date: 1/23/89

Industry/Toxicant: Gardinier
Address:
Contact/District:
Effluent Serial Number:
NPDES Permit Number:

BIOASSAY DATA FORM

Test Number: 2 of 3
Test Type: (Static) Renewal, (Screening) Conventional
Other:

Sample Collection: Date 8-8-88 Time 1041
Test Beginning: Date 8-9-88 Time
Test Ending: Date Time
Test Organism: Daphnia pulex
Age/Life Stage: neonate

Conc. or %	Test Container Number	Number of Live Organisms 0	24	48	72	96	Dissolved Oxygen (mg/l) 0	24	48	72	96	pH 0	24	48	72	96	Temperature °C 0	24	48	72	96	Conductivity 0	24	48	72	96
Control	26	10	10	10			8.9	8.6	8.9			8.2	7.9	8.0			19.9	19.4	19.3							
Control	132	10	10	9			8.9	8.7	8.9			8.2	8.0	8.1								205	223			
100%	41	10	10	9			7.1	8.8	1.8*			6.8	7.6	7.0			19.3	19.3	19.2							
100%	11	10	10	9			7.1	8.8	1.5*			6.8	7.6	7.0								2850	2450			
Initial of Investigator		KHL	RBF	MF			SHW	RBF	MF			SHW	RBF	MF			SHW	RBF	MF			SHW	RBF			

Alkalinity/Hardness (mg/l as $CaCO_3$)
Control Water: 118 120
100% Sample: 85 58
Other:
Measured by: MF

Ammonia unionized/total (mg/l as NH_3/N)
Hour 0
Control Water: 0
100% Sample: 2.89
Other:

Hour 0
Control Water:
100% Sample:
Other:
Measured by: MF

Signatures:
K.H. Lurding
Marshall Faircloth
Steven Wolfe
Russel Tryfaros

Instrument Calibrations:

	pH Meter:	DO Meter:	S-C-T Meter:
0 Hours			
24 Hours			
48 Hours			
72 Hours			
96 Hours			

Remarks: * checked 2X

Total Residual Chlorine:
Field data: Method:
Lab: Method:
Lab: Method:

Re-chlorinated:
Measurements by:

Reviewed by:
Date: 1-3-89

Gardiner

Test Number: 3 of 3
Test Type: (Static) Renewal Flow-through Recirculating
Other:

8-8-88 1041
8-9-88 1100
8-11-88 1100
Test Ending:
Test Organism: Notropis lee asi
Age/Life Stage: 17 days

Conc. or %	Test Container Number	% Mortality					Dissolved Oxygen (mg/l)					Temperature °C					Conductivity				
		0	24	48	72	96	0	24	48	72	96	0	24	48	72	96	0	24	48	72	96
Control	23	5	5	5			8.9	8.0	8.1			20.0	19.3	19.0							
Control	13	5	5	5			9.0	8.1	8.1								230	228			
100%	25	5	5	12			6.8	7.6	2.9*			19.2	19.4	19.1							
100%	5	6	6	2			6.8	7.6	3.2*								3090	3010			
Initial of Investigator		KHL	KHL	MF			SHW	RBF	MF			SHW	RBF	MF			SHW	MF		RBF	

Ammonia unionized/total (mg/l as NH3/N)
Hour 0

Control Water: 0
100% Sample: 2-89
Other:

Hour 0

Control Water:
100% Sample:
Other:
Measured by: MF

Control Water: 120 123
100% Sample: 85 58
Other:
Measured by: MF

Signatures:

K H Lurding
Marshall Fandel
Steven W Wolf
Russel Frydenborg

Instrument Calibrations:

	pH Meter:	DO Meter:	S-C-T Meter:
0 Hours			
24 Hours			
48 Hours			
72 Hours			
96 Hours			

Remarks: * checked 2x

Total Residual Chlorine:

Field data: _____ Method: _____
Lab: _____ Method: _____
Lab: _____ Method: _____

De-chlorinated:
Measurements by: _____

Reviewed by: [signature]

Date: 1/23/89

MICROTOX™ Data Reduction Sheet

SOURCE: Hardenner

GRAPH CALC EC 50 (15 MIN, 15 °C) = stimulatory %SAMPLE mg/L

SAMPLE DATA:

SAMPLE TYPE (EFFLUENT, LEACHATE, ETC.): effluent

DATE ASSAYED: 8-9-88 TIME: 10:00 AM

VISUAL COLOR: none CORRECTION REQUIRED: YES (NO)

VISUAL TURBIDITY: none SEPARATION REQUIRED: YES (NO)

INITIAL pH: pH ADJUSTED TO: WITH:

OSMOTICALLY ADJUSTED WITH: MOAS (DRY NaCl) OTHER

PRIMARY DILUTION OF SAMPLE:

OPERATOR: Leslie Williams REAGENT VIAL LOT NO. M-606

REMARKS:

MICROTOX™ 99% TEST

SAMPLE CUVETTE	% SAMPLE	READINGS 5 MIN	READINGS 15 MIN	Γ5	Γ15	NOTES
A1	0	73	50	X	X	
B1		72	52			
C1		70	44			
A2	12.38	83	58	-.126	-.156	
B2		80	58			
C2		83	57			
A3	24.75	83	71	-.192	-.324	
B3		90	70			
C3		93	75			
A4	49.5	90	71	-.215	-.330	
B4		92	73			
C4		92	74			
A5	99	91	76	-.221	-.384	
B5		90	79			
C5		95	82			
EC_{50} (MIN, °C)						

REVIEWED BY:

DATE: 1/23/89