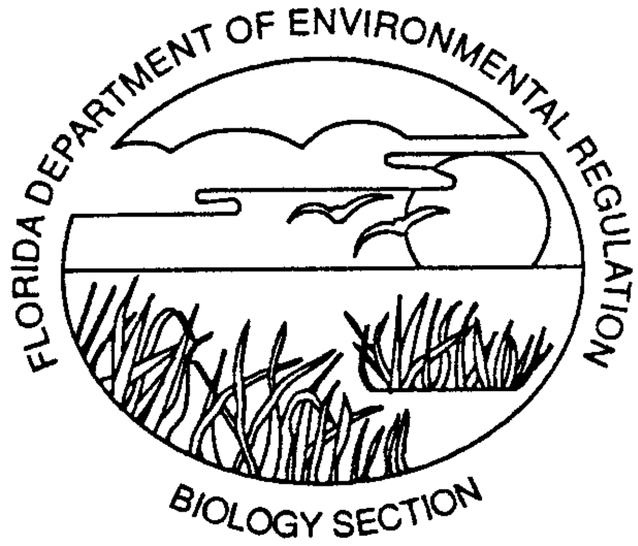


Bioassays of

Barefoot Bay Sewage Treatment Plant

Barefoot Bay, Brevard County, Florida

No NPDES Permit

Sampled 10/31/88

March 16, 1989

Biology Section
Division of Administrative and Technical Services

Bioassays of

Barefoot Bay Sewage Treatment Plant

Barefoot Bay, Brevard County, Florida

No NPDES Permit

Biology Section
Bureau of Laboratories
March 16, 1989

Executive Summary

Barefoot Bay Sewage Treatment Plant, Barefoot Bay, Brevard County, Florida, No NPDES Permit, on 1 to 3 November, 1988.

The Barefoot Bay Sewage Treatment Plant is a privately owned 1.0 MGD design flow contact stabilization STP. Discharge from the Plant is to a ditch to the Sebastian River then to the Indian River

The Biology Section performed six toxicity bioassays and two Microtox® assays on a sample of discharge from the facility. The Microtox® assays were performed on 1 November, 1988. The toxicity bioassays were performed on 1 to 3 November, 1988.

The sample of chlorinated discharge from the Barefoot Bay STP was toxic to the bioassay test organisms, *Ceriodaphnia dubia*, *Daphnia pulex*, and *Notropis leedsi*, producing LC_{50}'s of < 100%. The dechlorinated sample of discharge was toxic to the bioassay test organism, *C. dubia*, producing an LC_{50} of 92.86%. Both the chlorinated and dechlorinated samples adversely affected the Microtox® assay test organism, *Photobacterium phosphoreum*, producing EC_{50}'s of 8.17% and 43.47%, respectively.

The total residual chlorine concentration (0.8 mg/l Hach; 1.8 mg/l Iodometric) in the sample was sufficient to have caused the mortality noted in the chlorinated tests. Chemical analyses of the sample detected the pesticide, diazinon (0.025 to 0.05 µg/l) and several components sensitive to electron capture and nitrogen/phosphorus detection. The metals, arsenic (0.3 µg/l), silver (0.2 µg/l), and zinc (11.0 µg/l), were also detected in the sample. The diazinon was thought to have contributed to the deaths noted in the dechlorinated *C. dubia* tests.

In conclusion, the sample of discharge from the Barefoot Bay STP was toxic on 31 October, 1988.

Introduction

The Barefoot Bay Sewage Treatment Plant is located in Brevard County, Florida (Figure 1). This privately owned facility does not have a NPDES number. The plant is a 1.0 MGD design flow contact stabilization type sewage treatment plant with treatment consisting of extended aeration, aeration, clarification, chlorination, aerated sludge, and percolation and evaporation ponds (Figure 2). Discharge is to the Sebastian River then to the Indian River.

In response to a request form the FDER Central District office in Orlando, the Biology Section performed six (6) separate bioassays and two (2) Microtox® assays on a sample of discharge from the Barefoot Bay STP. The results of those 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.

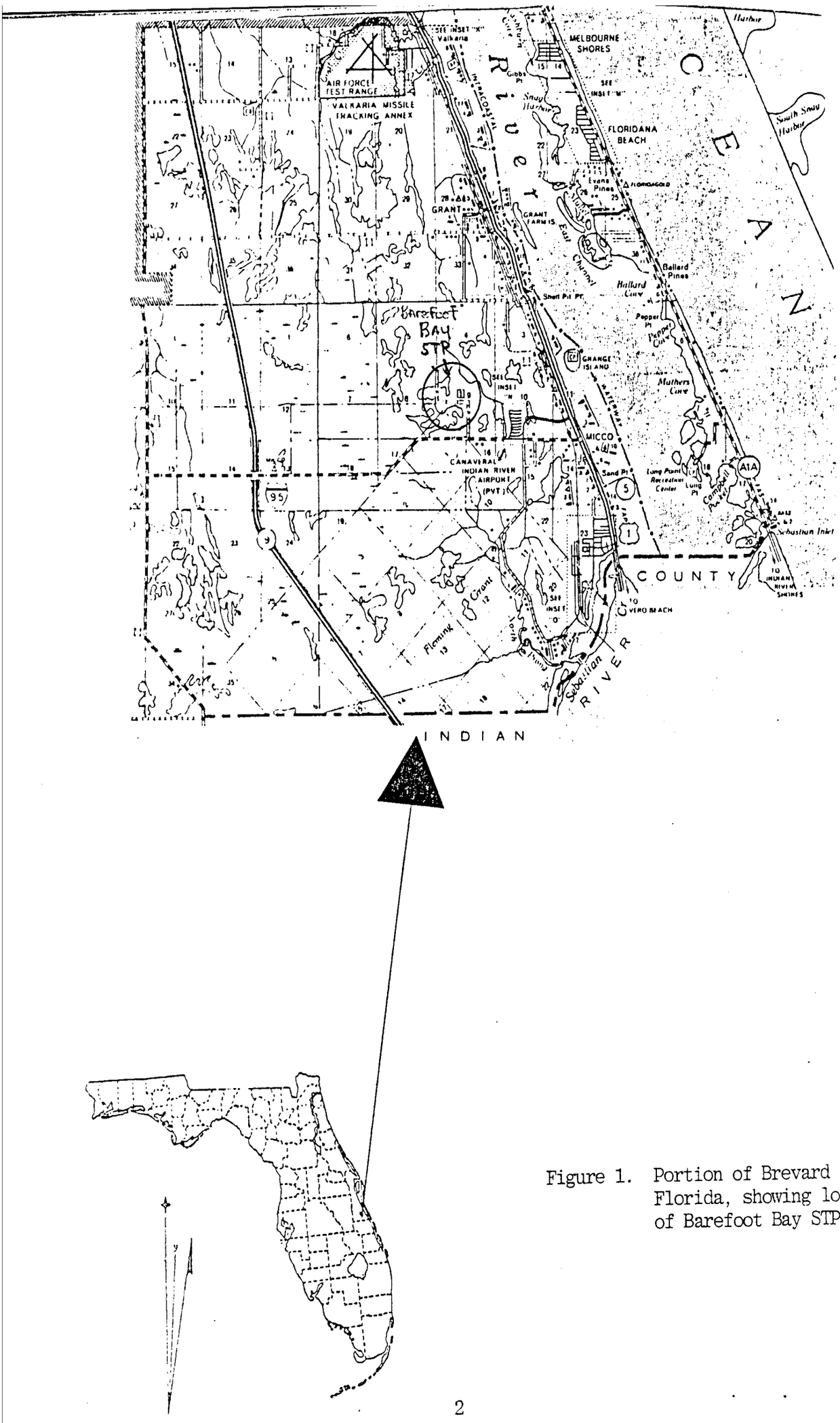


Figure 1. Portion of Brevard County Florida, showing location of Barefoot Bay STP.

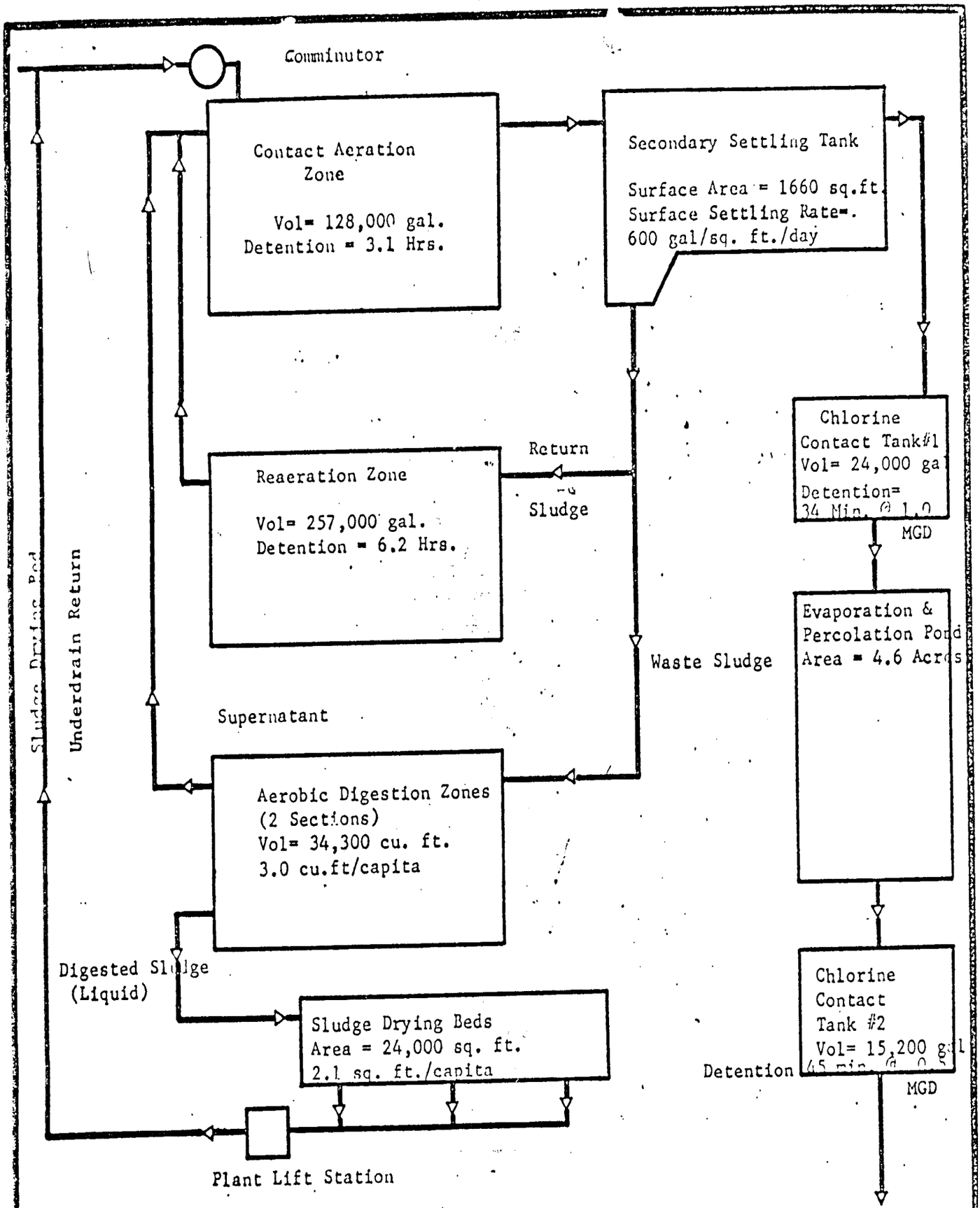


Figure 2.

GAC UTILITIES INC OF FLORIDA
BAREFOOT BAY DIVISION
SEWAGE TREATMENT PLANT
FLOW DIAGRAM

1.0 MGD - Contact Stabilization Type Sewage Treatment Plant (Diffused Air)

Prepared by: Henry B. Steeg & Associates, Inc.

November 11, 1971

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 10 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. Immediately prior to the assay, the culture is rehydrated with 1.0 ml of specially purified water (Microtox Reconstitution Solution).

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

Six separate 48 hour acute toxicity bioassays and two Microtox® assays were performed on a sample from the Barefoot Bay STP facility. The tests were performed from 1100 hours on 1 November to 1100 hours on 3 November, 1988.

Separate samples were obtained for chemical and biological testing. Samples for metal analyses were preserved with $HN0_3$. Samples for organic chemical analyses and for the bioassays were unpreserved. The samples were collected by Mr. David Herbster of the FDER Central District Office in Orlando at 1212 hours on 31 October, 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.

Total Residual Chlorine

Since many reclaimed waters are chlorinated, especially those originating from sewage treatment facilities, it is often desirable to differentiate between the toxicity resulting from chlorine and the toxicity associated with other components of the reclaimed water. To accomplish this, the Biology Section performed concurrent bioassays on a sample of chlorinated reclaimed water in which the chlorine had been neutralized. Dechlorination was accomplished by adding a calculated amount of 0.025N sodium thiosulfate. The total residual chlorine concentrations of the tested sample of reclaimed water are given on the bioassay data forms. A comparison of the results of these tests will distinguish between the toxicity due solely to residual chlorine and the toxicity inherently contained in the reclaimed water.

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 consisting of 30 ml plastic vials containing 15 mls of the control and each test concentration were used in bioassays with the water fleas, *Ceriodaphnia dubia* and *Daphnia pulex*. Adult *Ceriodaphnia* and *Daphnia* were separated using a nylon mesh sieve and placed into large culture dishes 18 hrs before the beginning of the test. Neonates produced during the intervening period (i.e., <18 hr old) were then captured by glass pipette, examined for injury, and transferred into the test chambers. Five individuals (for both *C. dubia* and *D. pulex*) were loaded

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 35 Conductance 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 Tables 2 and 3.

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 4.

Results and Discussion

The sample of chlorinated discharge from the Barefoot Bay Sewage Treatment Plant was toxic to the bioassay test organisms, the water fleas, *Ceriodaphnia dubia* and *Daphnia pulex*, and the fish, *Notropis leedsi*, producing LC_{50}'s of < 100%. The dechlorinated sample was toxic to the water flea, *C. dubia*, producing an LC_{50} of 92.86%. Both the chlorinated and dechlorinated samples adversely affected the Microtox® assay organism, *Photobacterium phosphoreum*, producing EC_{50}'s of 8.17% and 43.47%, respectively.

The total residual chlorine concentration (0.8 mg/l Hach; 1.8 mg/l Iodometric) was sufficient to have caused the mortality in the chlorinated sample. The total ammonia (4.20 mg/l) and unionized ammonia (0.07 mg/l) probably did not contribute to the toxicity of the sample. Chemical analyses detected the pesticide, diazinon (0.025 to 0.05 µg/l) and several unidentified components sensitive to ECD and NPD detection in the organics sample. The metals, arsenic (0.3 µg/l), silver (0.2 µg/l), and zinc (11 µg/l), were also detected in the sample.

The diazinon may have contributed to the observed deaths in the dechlorinated sample. Diazinon concentrations of 0.05 µg/l have produced toxicity in tests with *C. dubia* (personal communication D. Mount, U.S. EPA, Duluth).

The sample of discharge from the Barefoot Bay STP was toxic on 31 October, 1988. The toxicity was attributed to chlorine and the pesticide diazinon.

Biology Section Recommendations

The Biology Section recommends that, if this facility will be discharging to surface waters at any time, the facility be required to undertake additional toxicity testing of their discharge as the first step in a Toxicity Reduction Evaluation. Should additional toxicity tests show that the discharge from the facility is again toxic, then the facility should be required to continue the Toxicity Reduction Evaluation with the goal of meeting state water quality standards.

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
10.0%	13.5 ml	1.5 ml	15 ml
30.0%	10.5 ml	4.5 ml	15 ml
50.0%	7.5 ml	7.5 ml	15 ml
70.0%	4.5 ml	10.5 ml	15 ml
85.0%	2.25 ml	12.75 ml	15 ml
100.0%	0.0 ml	15.0 ml	15 ml

Daphnia pulex (water flea)

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

Notropis leedsi (bannerfin shiner)

Concentration	Dilution Water	Sample	Total Volume
Control	500 ml	0 ml	500 ml
10.0%	450 ml	50 ml	500 ml
30.0%	350 ml	150 ml	500 ml
50.0%	250 ml	250 ml	500 ml
70.0%	150 ml	350 ml	500 ml
85.0%	75 ml	425 ml	500 ml
100.0%	0 ml	500 ml	500 ml

Table 2. Data recorded during the 48 hour static bioassay of a chlorinated sample of discharge from outfall #001 of the Barefoot Bay STP, No NPDES #, Brevard County, Florida, on 1 November to 3 November, 1988.

Facility:	**Barefoot Bay STP**		**NPDES #**	**None**	Facility Type: **Private**
Location:	-		Contact/District:	D. Herbster / SE	
County:	Brevard		Test type:	Static Screen	
Sample Collection Date:	**10/31/88**	Time: 1212	# tests:	6	Receiving Water: ditch to Sebastian River to Indian River
Test Beginning Date:	11/1/88	Time: 1100	Chlorination Type:	**Chlorinated**	
Test Ending Date:	11/3/88	Time: 1100			

Total Residual Cl2	mg/l	Method
Field:	-	-
Lab:	0.8	Hach
Lab:	1.8	Iodometric

Ammonia	Total (mg/l)	Unionized (mg/l)
Control Water (fish):	0.00	0.00
Cond. Control Water (daphnids):	0.00	0.00
100% Sample:	4.20	0.07

Alk & Hardness	Alkalinity (mg/l)	Hardness (mg/l)
Control Water (fish):	125	122
Cond. Control Water (daphnids):	130	135
100% Sample:	160	150

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

Organism: ***Ceriodaphnia dubia*** Life stage: neonate
LC50: <100%

Concentrations	SURVIVAL # at 0 hr	Live at 24 hr	Live at 48 hr	Dissolved Oxygen 0 hour	24 hour	48 hour	pH 0 hour	24 hour	48 hour	Temperature 0 hour	24 hour	48 hour	Conductivity 0 hour	48 hour
Control	5	5	-	8.0	8.4	-	8.4	8.0	-	19.3	20.5	-	-	-
Control	5	5	-	8.0	8.5	-	8.4	8.0	-	-	-	-	-	-
Control	5	5	-	8.2	8.5	-	8.3	8.0	-	-	-	-	-	-
Control	5	5	-	8.3	8.5	-	8.4	8.0	-	-	-	-	220	245
100%	5	0	-	7.4	8.8	-	7.3	8.1	-	19.6	20.7	-	-	-
100%	5	0	-	7.5	8.8	-	7.3	8.1	-	-	-	-	-	-
100%	5	0	-	7.4	8.9	-	7.3	8.1	-	-	-	-	-	-
100%	5	0	-	7.4	8.9	-	7.6	8.1	-	-	-	-	855	870

Organism: ***Daphnia pulex*** Life stage: neonate
LC50: <100%

Concentrations	SURVIVAL # at 0 hr	Live at 24 hr	Live at 48 hr	Dissolved Oxygen 0 hour	24 hour	48 hour	pH 0 hour	24 hour	48 hour	Temperature 0 hour	24 hour	48 hour	Conductivity 0 hour	48 hour
Control	5	5	-	8.3	8.4	-	8.4	8.0	-	19.0	20.8	-	-	-
Control	5	5	-	8.4	8.4	-	8.4	8.0	-	-	-	-	-	-
Control	5	5	-	8.4	8.4	-	8.4	8.0	-	-	-	-	-	-
Control	5	5	-	8.4	8.4	-	8.4	8.0	-	-	-	-	220	255
100%	5	0	-	7.4	8.8	-	7.3	8.1	-	19.2	20.8	-	-	-
100%	5	0	-	7.5	8.8	-	7.3	8.1	-	-	-	-	-	-
100%	5	0	-	7.4	8.8	-	7.3	8.1	-	-	-	-	-	-
100%	5	0	-	7.4	8.8	-	7.3	8.1	-	-	-	-	865	920

Organism: ***Notropis leedsi*** Age: 10 days
LC50: <100%

Concentrations	SURVIVAL # at 0 hr	Live at 24 hr	Live at 48 hr	Dissolved Oxygen 0 hour	24 hour	48 hour	pH 0 hour	24 hour	48 hour	Temperature 0 hour	24 hour	48 hour	Conductivity 0 hour	48 hour
Control	5	5	-	8.0	8.8	-	7.8	7.9	-	20.7	19.5	-	-	-
Control	5	5	-	8.0	8.8	-	7.8	7.9	-	-	-	-	225	220
100%	5	0	-	6.7	8.6	-	7.2	7.6	-	21.0	19.6	-	-	-
100%	5	0	-	6.6	8.6	-	7.2	7.6	-	-	-	-	935	910

Table 3. Data recorded during the 48 hour static definitive bioassay of a dechlorinated sample of discharge from outfall #001 of the Barefoot Bay STP, No NPDES #, Brevard County, Florida, on 1 to 3 November, 1988.

Facility: **Barefoot Bay STP** NPDES # **None** Facility Type: **Private**
Location: Barefoot Bay Contact/District: D. Herbster / SE
County: Brevard Test type: definitive
Sample Collection Date: **10/31/88** Time: 1212 # tests: 6 Receiving Water: ditch to Sebastian River
Test Beginning Date: 11/1/88 Time: 1100 Chlorination Type: **Dechlorinated** to Indian River
Test Ending Date: 11/3/88 Time: 1100 **Page 1 of 2**

Total Residual Cl2	mg/l	Method
Field:	-	-
Lab:	0.8	Hach
Lab:	1.8	Iodometric

Ammonia	Total (mg/l)	Unionized (mg/l)
Control Water (fish):	0.00	0.00
Cond. Control Water (daphnids):	0.00	0.00
100% Sample:	4.20	0.07

Alk & Hardness	Alkalinity (mg/l)	Hardness (mg/l)
Control Water (fish):	125	122
Cond. Control Water (daphnids):	130	135
100% Sample:	160	150

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

Organism: ***Ceriodaphnia dubia*** Life stage: neonate
LC50: - Binomial: 92.9% Moving Average: 92.9% 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	8.5	-	8.4	8.4	-	8.3	19.8	-	21.1	-	-
Control	5	5	4	8.5	-	8.5	8.4	-	8.3	-	-	-	-	-
Control	5	5	4	8.5	-	8.5	8.4	-	8.3	-	-	-	-	-
Control	5	5	5	8.5	-	8.5	8.4	-	8.3	-	-	-	220	260
10%	5	5	5	8.5	-	8.6	8.2	-	8.3	19.5	-	21.1	-	-
10%	5	5	5	8.6	-	8.6	8.2	-	8.3	-	-	-	-	-
10%	5	5	5	8.6	-	8.6	8.2	-	8.3	-	-	-	-	-
10%	5	5	5	8.6	-	8.6	8.2	-	8.3	-	-	-	295	320
30%	5	5	5	8.4	-	8.6	7.9	-	8.3	19.8	-	21.2	-	-
30%	5	5	5	8.4	-	8.6	7.9	-	8.3	-	-	-	-	-
30%	5	5	5	8.4	-	8.6	7.9	-	8.2	-	-	-	-	-
30%	5	5	5	8.4	-	8.6	7.9	-	8.2	-	-	-	730	460
50%	5	5	5	8.3	-	8.6	7.8	-	8.2	19.8	-	21.0	-	-
50%	5	5	5	8.3	-	8.6	7.8	-	8.1	-	-	-	-	-
50%	5	5	5	8.3	-	8.6	7.8	-	8.1	-	-	-	-	-
50%	5	5	5	8.4	-	8.6	7.8	-	8.1	-	-	-	560	600
70%	5	5	5	8.3	-	8.6	7.8	-	8.1	20.2	-	21.0	-	-
70%	5	5	5	8.3	-	8.6	7.8	-	8.1	-	-	-	-	-
70%	5	5	5	8.3	-	8.7	7.8	-	8.1	-	-	-	-	-
70%	5	5	5	8.3	-	8.7	7.8	-	8.1	-	-	-	695	735
85%	5	5	5	8.1	-	8.6	7.7	-	8.1	20.4	-	21.0	-	-
85%	5	5	4	8.1	-	8.6	7.7	-	8.0	-	-	-	-	-
85%	5	5	5	8.1	-	8.6	7.7	-	8.0	-	-	-	-	-
85%	5	5	5	8.2	-	8.6	7.7	-	8.0	-	-	-	800	860
100%	5	5	1	7.8	-	8.6	7.5	-	8.0	20.5	-	21.0	-	-
100%	5	5	0	7.9	-	8.6	7.5	-	8.0	-	-	-	-	-
100%	5	5	1	7.9	-	8.6	7.5	-	8.0	-	-	-	-	-
100%	5	5	0	8.0	-	8.6	7.5	-	8.0	-	-	-	910	975

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	5	5	8.3	-	8.5	8.4	-	8.2	19.7	-	21.0	-	-
Control	5	5	5	8.4	-	8.5	8.4	-	8.2	-	-	-	-	-
Control	5	5	5	8.4	-	8.6	8.4	-	8.2	-	-	-	-	-
Control	5	5	5	8.4	-	8.6	8.4	-	8.2	-	-	-	220	255
100%	5	5	5	8.1	-	8.6	7.5	-	8.0	19.9	-	19.9	-	-
100%	5	5	5	8.1	-	8.6	7.5	-	8.0	-	-	-	-	-
100%	5	5	5	8.0	-	8.6	7.5	-	8.0	-	-	-	-	-
100%	5	5	5	8.0	-	8.6	7.5	-	8.0	-	-	-	870	960

Table 3. Continued.

Facility:	**Barefoot Bay STP**		**NPDES # None**	Facility Type: **Private**	
Location:	Barefoot Bay		Contact/District: D. Herbster / SE		
County:	Brevard		Test type: definitive		
Sample Collection Date:	**10/31/88**	Time: 1212	# tests: 6	Receiving Water: ditch to Sebastian River to Indian River	
Test Beginning Date:	11/1/88	Time: 1100	Chlorination Type: **Dechlorinated**		
Test Ending Date:	11/3/88	Time: 1100		**Page 2 of 2**	

Total Residual Cl2	mg/l	Method
Field:	-	-
Lab:	0.8	Hach
Lab:	1.8	Iodometric

Ammonia	Total (mg/l)	Unionized (mg/l)
Control Water (fish):	0.00	0.00
Cond. Control Water (daphnids):	0.00	0.00
100% Sample:	4.20	0.07

Alk & Hardness	Alkalinity (mg/l)	Hardness (mg/l)
Control Water (fish):	125	122
Cond. Control Water (daphnids):	130	135
100% Sample:	160	150

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

Organism: ***Notropis leedsi*** Age: 10 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	8.0	8.8	8.2	7.9	8.0	8.0	20.8	20.1	20.1	-	-
Control	5	5	5	8.1	8.7	8.2	7.9	8.0	8.0	-	-	-	225	225
10%	5	5	5	8.0	8.8	8.0	7.7	7.9	7.9	20.8	19.8	19.7	-	-
10%	5	4	4	8.0	8.8	8.0	7.7	7.9	7.9	-	-	-	295	290
30%	5	5	5	8.0	8.7	7.9	7.6	7.9	7.9	20.9	19.5	19.6	-	-
30%	5	5	5	8.0	8.7	7.9	7.6	7.9	7.9	-	-	-	445	430
50%	5	5	5	7.8	8.7	7.6	7.5	7.8	7.8	21.0	19.4	19.5	-	-
50%	5	4	4	7.8	8.7	7.7	7.5	7.8	7.8	-	-	-	590	570
70%	5	5	5	7.4	8.4	7.7	7.3	7.7	7.8	21.0	19.3	19.4	-	-
70%	5	5	5	7.5	8.5	7.7	7.3	7.7	7.8	-	-	-	730	710
85%	5	5	5	7.1	8.5	7.8	7.2	7.6	7.8	21.1	19.3	19.4	-	-
85%	5	5	5	7.1	8.6	7.8	7.3	7.7	7.8	-	-	-	845	820
100%	5	5	5	6.8	8.5	7.8	7.2	7.6	7.8	20.4	19.5	19.5	-	-
100%	5	5	5	6.8	8.5	7.8	7.2	7.6	7.8	-	-	-	955	920

Table 4. Microtox test results, Barefoot Bay STP discharge, Brevard County, no NPDES #, on 1 November, 1988.

Microtox Test Culture Lot # M-606

% Sample	Final Reclaimed Water			
	Chlorinated		Dechlorinated	
	5-min	15-min	5-min	15-min
0	81	54	70	46
0	77	50	72	47
0	78	50	70	44
12.38	48	26	60	37
12.38	45	25	61	38
12.38	52	29	61	39
24.75	27	16	55	32
24.75	27	16	52	32
24.75	27	16	53	32
49.5	9	5	37	21
49.5	9	5	36	21
49.5	9	5	34	20
99	0.5	0.3	21	12
99	0.5	0.3	19	11
99	0.5	0.3	19	11

EC_{50} Chlorinated (15 minute, 15 °C) = 8.17% sample
EC_{50} Dechlorinated (15 minute, 15 °C) =43.47% sample

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. LC_{50} Data for Bioassay

Organism	Method of Data Analysis	48 Hour LC_{50} (95% Confidence Intervals)
Chlorinated discharge		
Ceriodaphnia dubia	noncalculated	<100% [a]
Daphnia pulex	noncalculated	<100% [a]
Notropis leedsi	noncalculated	<100% [a]
Dechlorinated discharge		
Ceriodaphnia dubia	Binomial Moving Average Probit	92.86% (85.00%, 100.00%) 92.86% (90.40%, 95.54%) [b]
Daphnia pulex	noncalculated	not toxic
Notropis leedsi	noncalculated	not toxic

a The LC_{50} is < 100% since all the organisms died in the lowest test concentration (100%).

b Due to an insufficient number of dead organisms, or to the irregular distribution of mortality, an LC_{50} value cannot be calculated using this method of statistical analysis.

Table 6. Results of chemical analyses on the sample of discharge from the Barefoot Bay STP, Brevard County, Florida.

Organics

Acid Extractables	None detected
Base Neutral Extractables	None detected
Pesticide Extractables	Diazinon (0.05µg/l)[k]
	Several unidentified components sensitive to ECD and NPD

Metals

Arsenic	0.3 µg/l
Silver	0.2 µg/l
Zinc	11.0 µg/l

[k] actual value is known to be less than value given, but greater than half this value.

Literature Cited

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

Florida Department of Environmental Regulation. 1985. Quality assurance manual for performing acute toxicity tests (Second Edition). Biology Section, Tallahassee.

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.

Discussion of the Toxicity Criteria

The toxicity standards of Chapters 17-3 and 17-4, F.A.C., are complex. Because of this complexity, they will be briefly reviewed here so that these test results may be more readily compared with the standards. For legal purposes, however, the language of the Florida Administrative Code should be consulted directly. Applicable portions of the F.A.C. are: 17-3.021(1), 17-3.021(4), 17-3.051(1)(d), 17-3.061(3)(p), 17-3.402(1), 17-4.244(4), and 17-4.244(6).

The particular restrictions applied to a given discharger depend upon the type of discharge, the type of water body which is impacted, and the type of mixing zone, if any, which has been permitted. The most stringent toxicity limitations which apply to the various combinations of these factors are listed below, specified in terms of the most dilute concentration of effluent which is allowed to kill 50% of the test organisms in 96 hours. Note that any listed concentration greater than 100% is extrapolated from toxicity curves determined from testing various effluent concentrations of 100% or less. Violations of the toxicity standards occur in the following conditions.

I. Discharges to Surface Water

   A. With no mixing zone, or with only a dredge and fill mixing zone the discharge may not exhibit an $LC_{50} < 2000\%$ at the point of discharge.

   B. With a non-dredge and fill mixing zone the discharge may not exhibit an $LC_{50} < 100\%$ at the point of discharge and may not exhibit an $LC_{50} < 2000\%$ at the edge of the mixing zone.

   C. With a non-dredge and fill mixing zone, and either discharging to the open ocean through multiport diffusers or meeting the provisions of 17-4.244(4)(b) the LC_{50} at the point of discharge cannot exceed 30% sample concentration.

II. Discharges to ground water may not exceed an LC_{50} of 300% at the point of contact with surface waters

A violation of these provisions occurs if toxicity is found at a more dilute concentration of the sampled discharge than allowed above.

Industry/Toxicant Barefoot Bay STP
Address Brevard Co.
Contact/District
Effluent Serial Number
NPDES Permit Number NONE

BIOASSAY DATA FORM

Test Number: 1 of 6
Test Type: Static Renewal, Screening Conventional
Other: de Cl2

Sample Collection: Date 10-31-88 Time 12:12
Test Beginning: Date 11-1-88 Time 11:00
Test Ending: Date 11-3-88 Time 11:00
Test Organism Ceriodaphnia dubia
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	1/2	5/5	5/5	5/4			8.5/8.5	—	8.4/8.5			8.4/8.4	—	8.3/8.3			14.8/	—	21.1/21.4	5C						
Control	3/4	5/5	5/5	4/5			8.5/8.5	—	8.5/8.5			8.4/8.4	—	8.3/8.3								220	—	260		
10%	5/6	5/5	5/5	5/5			8.5/8.6	—	8.6/8.6			8.2/8.2	—	8.3/8.3			19.5/	—	21.1/							
10%	7/8	5/5	5/5	5/5			8.6/8.6	—	8.6/8.6			8.2/8.2	—	8.3/8.3								295	—	320		
30%	9/10	5/5	5/5	5/5			8.4/8.4	—	8.6/8.6			7.9/7.9	—	8.3/8.2			19.8/	—	21.2/							
30%	11/12	5/5	5/5	5/5			8.4/8.4	—	8.6/8.6			7.9/7.9	—	8.2/8.2								430	—	460		
50%	13/14	5/5	5/5	5/5			8.3/8.3	—	8.4/8.4			7.8/7.8	—	8.1/8.1			19.8/	—	21.0/							
50%	15/16	5/5	5/5	5/5			8.3/8.4	—	8.6/8.6			7.8/7.8	—	8.1/8.1								560	—	600		
70%	17/18	5/5	5/5	5/5			8.3/8.3	—	8.4/8.4			7.8/7.8	—	8.1/8.1			20.2/	—	21.0/							
70%	19/20	5/5	5/5	5/5			8.3/8.3	—	8.7/8.7			7.8/7.8	—	8.1/8.1								695	—	735		
85%	21/22	5/5	5/5	5/4			8.1/8.1	—	8.6/8.6			7.7/7.7	—	8.0/8.0			20.4/	—	21.0/							
85%	23/24	5/5	5/5	5/5			8.1/8.4	—	8.6/8.6			7.7/7.7	—	8.0/8.0								800 KLH		860		
100%	25/26	5/5	5/5	1/0			7.8/7.9	—	8.6/8.6			7.5/7.5	—	8.0/8.0			20.5/	—	21.0/							
100%	27/28	5/5	5/5	1/0			7.4/8.0	—	8.6/8.6			7.5/7.5	—	8.0/8.0								1110	—	975		
Initial of Investigator		JHW	JHW	MF			SHW		JMF			SHW		JMF			SHW		JMF			SHW		JMF		

Alkalinity/Hardness (mg/l as $CaCO_3$)
Control Water: 130/135
100% Sample: 160/150
Well ~~Other~~: 125/122
Measured by: JMF

Ammonia unionized/total (mg/l as NH_3/N)
Hour 0
Control Water: 0.0
100% Sample: 4.2
well Other: 0.0

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

Slope
-59.0
New membrane installed

Signatures:
Marshall Faudale
Kelly Wardina
Steven Wolfe

Instrument Calibrations:

	pH Meter:	DO Meter:	S-C-T Meter:
0 Hours	7.0, 9.0	9.0 @ 21°C	1270 @ 1294 @ 20.8°C
24 Hours	7.0, 9.0	9.0 @ 21°C	1291 & 1322 @ 21.1
48 Hours	7.0, 9.1	9.0 @ 20°C	1285 @ 1285 @ 20.8
72 Hours			
96 Hours			

Total Residual Chlorine:
Field data: — Method: —
Lab: 0.8 Method: Hach
Lab: 1.17 Method: EPA

Reviewed by:
Date: 3/13/89

Remarks: 48h DO 21.1°C T 20.9
24h DO 19.7 T 19.7°C
48h DO 19.5°C T 19.5°C

Dechlorinated:
Measurements by: MF

Industry/Facility: Barefoot Bay STP
Address:
Contact/District:
Effluent Serial Number:
NPDES Permit Number:

Test Number: 2 of 6
Test Type: Static Renewal, Screening Conventional
Other: deCl2

Sample collection: Date 10-31-88 Time 10:12
Test beginning: Date 11-1-88 Time 11:00
Test Ending: Date 11-3-88 Time 11:00
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	29	5	5	5			8.3	—	8.5			8.4	—	8.2			19.7	—	21.0							
"	30	5	5	5			8.4	—	8.5			8.4	—	8.2												
"	31	5	5	5			8.4	—	8.6			8.4	—	8.2												
"	32	5	5	5			8.4	—	8.6			8.4	—	8.2								220	—	255		
100%	33	5	5	5			8.1	—	8.6			7.5	—	8.0			19.9	—	19.9							
"	34	5	5	5			8.1	—	8.4			7.5	—	8.0												
"	35	5	5	5			8.0	—	8.4			7.5	—	8.0												
"	36	5	5	5			8.0	—	8.6			7.5	—	8.0								870	—	960		
Initial of Investigator		JUW	SHW	MJF			SHW		JMF			SHW		JMF			SHW		JMF			SHW		JMF		

Alkalinity/hardness (mg/l as $CaCO_3$)
Control Water: 130/135
100% Sample: 160/150
Other:
Measured by: JMF

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

Hour _____
Control Water:
100% Sample:
Other:
Measured by: _____

Signatures:
Marshall Faircloth
K M Lurding
Steven H Wolfe

Instrument Calibrations:

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

Remarks:

Total Residual Chlorine:
Field date: _____ Method: _____
Lab: _____ Method: _____
Lab: _____ Method: _____

Reviewed by:
Date: 3/13/87

Data entered/verified:
Data entered/verified by:

Industry/Toxicant: Barefoot Bay STP
Address:
Contact/District:
Effluent Serial Number:
NPDES Permit Number:

BIOASSAY DATA FORM

Test Number: 3 of 6
Test Type: Static Renewal, Screening Conventional
Other: de Cl2

Sample Collection: Date 10-31-88 Time 12:12
Test Beginning: Date 11-1-88 Time 11:00
Test Ending: Date 11-3-88 Time 11:00
Test Organism: Notropis leedsi
Age/Life Stage: 10 days

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	38	5	5	5			8.0	8.8	8.2			7.9	8.0	8.0			20.8	20.1	20.1							
Control	100	5	5	5			8.1	8.7	8.2			7.9	8.0	8.0								225	–	225		
10%	5	5	5	5			8.0	8.8	8.0			7.7	8.0	7.9			20.8	19.8	19.7							
10%	113	4	4	4			8.0	8.8	8.0			7.7	8.0	7.9								295	–	290		
30%	39	5	5	5			8.0	8.7	7.9			7.6	7.9	7.9			20.9	19.5	19.6							
30%	72	5	5	5			8.0	8.7	7.9			7.6	7.9	7.9								445	–	430		
50%	99	5	5	5			7.8	8.7	8.6	7.6		7.5	7.8	7.8			21.0	19.4	19.5							
50%	21	5	4	4			7.8	8.7	8.7	7.7		7.5	7.8	7.8								590	–	570		
70%	44	5	5	5			7.4	8.4	7.7			7.3	7.7	7.8			21.0	19.3	19.4							
70%	6	5	5	5			7.5	8.5	7.7			7.3	7.7	7.8								730	–	710		
85%	34	5	5	5			7.1	8.5	7.8			7.2	7.6	7.8			21.1	19.3	19.4							
85%	18	5	5	5			7.1	8.6	7.8			7.3	7.7	7.8								845	–	820		
100%	49	5	5	5			6.8	8.5	7.8			7.2	7.6	7.8			20.4	19.5	19.5							
100%	130	5	5	5			6.8	8.5	7.8			7.2	7.6	7.8								955	–	920		
Initial of Investigator		KHL	AHW	MH			SHW	JMF	JMF			SHW	JMF	JMF			SHW	JMF	SHW			SHW		JMF		

Alkalinity/Hardness (mg/l as $CaCO_3$)
Control Water: 125/122
100% Sample: 160/150
Other:
Measured by: JMF

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

Signatures:
M. Brandt
K M Lurding
Steven H Wolfe

Instrument Calibrations:

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

Remarks:

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

Dechlorinated:
Measurements by:

Reviewed by:
Date: 3/13/89

Facility/Source: Barefoot Bay STP
Address:
Contact/District:
Effluent Serial Number:
NPDES Permit Number:

BIOASSAY DATA SHEET

Test Number: 4 of 6
Test Type: Static Renewal, Screening Conventional
Other: Clz

Sample Collection: Date 10-31-88 Time 12:12
Test Beginning: Date 11-1-88 Time 11:00
Test Ending: Date 11-3-88 Time 11:00
Test Organism: C. dubia
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	37	5	5				8.0	8.4				8.4	8.0				19.3	20.5	~~20.1~~ SW							
"	38	5	5				8.0	8.5				8.4	8.0													
"	39	5	5				8.2	8.5				8.3	8.0													
"	40	5	5				8.3	8.5				8.4	8.0									220	245			
100%	41	5	0				7.4	8.8				~~8.3~~ 8.3	8.1				~~19.2~~ 19.1	20.7								
"	42	5	0				7.5	8.9				7.3	8.1													
"	43	5	0				7.4	8.9				7.3	8.1													
"	44	5	0				7.4	8.9				7.3	8.1									855	870			
Initial of Investigator		SHW	JHF				SHW	JHF				SHW	JHF				SHW	JHF				SHW	JHF			

Alkalinity/Hardness (mg/l as CaCO3)
Control Water:
100% Sample:
Other:
Measured by:

Ammonia unionized/total (mg/l as NH3/N)
Hour 0:
Control Water:
100% Sample:
Other:
Hour
Control Water:
100% Sample:
Other:
Measured by:

Signatures: M. Faucett, K M Lurding, Steven W Wolfe

Instrument Calibrations:

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

Remarks:

Total Residual Chlorine:
Field data: Method:
Lab: Method:
Lab: Method:
Reconciliated:
Data entered by:

Reviewed by:
Date: 3/13/89

10-31-88 12:12

Industry/Toxicant: Barefoot Bay STP
Address:
Contact/District:
Effluent Serial Number:
NPDES Permit Number:

BIOASSAY DATA FORM

Test Number: 5 of 6
Test Type: Static Renewal, Screening Conventional
Other: Cl2

Sample Collection: Date 10-31 Time 8:00
Test Beginning: Date 11-1-88 Time 11:00
Test Ending: Date 11-3-88 Time 11:00
Test Organism: D. 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	45	5	5				8.3	8.4				8.4	8.0				19.0	20.8								
"	46	5	5				8.4	8.4				8.4	8.0													
"	47	5	5				8.4	8.4				8.4	8.0													
"	48	5	5				8.4	8.4				8.4	8.0									220	255			
100%	49	5	0				7.4	8.8				7.3	8.1				19.2	20.8								
"	50	5	0				7.5	8.8				7.3	8.1													
"	51	5	0				7.4	8.8				7.3	8.1													
"	52	5	0				7.4	8.8				7.3	8.1									865	920			
Initial of Investigator		JW	JW				SW	JMF				SW	JMF				SW	JMF				SW	JMF			

Alkalinity/Hardness (mg/l as $CaCO_3$)

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

Ammonia unionized/total (mg/l as NH_3/N)
Hour 0

Control Water:
100% Sample:
Other:

Hour

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

Signatures:
M Fauchett
KA Lurding
Steven H Wolfe

Instrument Calibrations:

	pH Meter:	D.O. Meter:	S-C-T Meter:
0 Hours			
24 Hours			
48 Hours			
72 Hours			
96 Hours			

Remarks:

Total Residual Chlorine:

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

Dechlorinated:
Measurements by:

Reviewed by:
Date: 3/13/89

Industry/Major: Barefoot Bay STP
Address:
Contact/District:
Effluent Serial Number:
NPDES Permit Number:

BIOASSAY DATA FORM

Test Number: 6 of 6
Test Type: Static Renewal, Screening Conventional
Other: Cl2

Sample Collection: Date 10-31-88 Time 12:12
Test Beginning: Date 11-1-88 Time 1100
Test Ending: Date 11-3-88 Time 1100
Test Organism: N. leptodai
Age/Life Stage: 10 days

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	18	5	5				8.0	8.8				7.8	7.9				20.7	19.5								
Control	9	5	5				8.0	8.8				7.8	7.9									225	220			
100%	31	5	0				6.7	8.6				7.2	7.6				21.0	19.6								
100%	47	5	0				6.6	8.6				7.2	7.6									935	910			
Initial of Investigator		KML	SHW				SHW/JMF					SHW/JMF					SHW/JMF					SHW/JMF				

Alkalinity/Hardness (mg/l as $CaCO_3$)
Control Water:
100% Sample:
Other:
Measured by:

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

Signatures:
M. Faulth
K. M. Harding
Steven H. Way

Instrument Calibrations:

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

Remarks:

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

Re-chlorinated:
Monitoring done by:

Reviewed by:
Date: 3/13/89

MICROTOX™ Data Reduction Sheet

SOURCE: Barefoot Bay STP

GRAPH CALC EC 50 (15 MIN, 15 °C) = 8.17 (%SAMPLE) mg/L

SAMPLE DATA:

SAMPLE TYPE (EFFLUENT, LEACHATE, ETC.): effluent Cl2

DATE ASSAYED: 11-1-88 TIME: 9:30 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	81	54	X	X	
B1		77	50			
C1		78	50			
A2	~~12.38~~ 6.19	48	26	.628	.925	
B2		45	25			
C2		52	29			
A3	~~24.75~~ 12.38	27	16	1.914	2.208	
B3		27	16			
C3		27	16			
A4	~~49.5~~ 24.75	9	5	14.733	9.267	
B4		9	5			
C4		9	5			
A5	~~99~~ 49.5	.5	.3	136.33	170.11	
B5		.5	.3			
C5		.5	.3			
EC_{50} (MIN, °C)				8.51	8.17	

REVIEWED BY:

DATE: 3/13/89

MICROTOX™ Data Reduction Sheet

SOURCE: Barefoot Bay STP

GRAPH CALC EC 50 (15 MIN, 15 °C) = 43.47 (%SAMPLE) mg/L

SAMPLE DATA:

SAMPLE TYPE (EFFLUENT, LEACHATE, ETC.): effluent de Cl2

DATE ASSAYED: 11-1-88 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	70	46	X	X	
B1		72	47			
C1		70	44			
A2	12.38	60	37	.165	.202	
B2		61	38			
C2		61	39			
A3	24.75	55	32	.325	.427	
B3		52	32			
C3		53	32			
A4	49.5	37	21	.981	1.210	
B4		36	21			
C4		34	20			
A5	99	21	12	2.593	3.029	
B5		19	11			
C5		19	11			
EC_{50} (MIN, ° C)				50.45	43.47	

REVIEWED BY:

DATE: 3/13/89