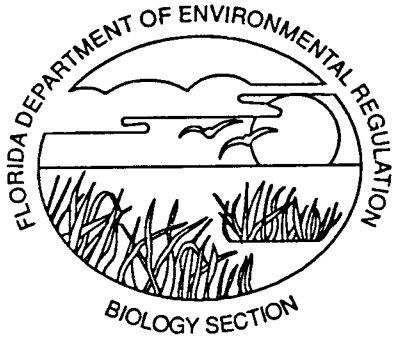


Bioassays of

Bay CountyWastewater Treatment Plant

Panama City, Bay County, Florida

NPDES #FL0002631

Sampled 6/6/88

September 22, 1988

Biology Section
Division of Administrative and Technical Services

Bioassays of

Bay County Wastewater Treament Plant

Panama City, Bay County, Florida

NPDES #FL0002631

Biology Section
Bureau of Laboratories
September 22, 1988

Executive Summary

Bay County Wastewater Treatment Plant, NPDES #FL0002631, on 7 to 9 June, 1988.

The sample collected from the Lagoon Weir of the Bay County WWTP was not acutely toxic to the bioassay test organisms, *Ceriodaphnia dubia*, *Daphnia pulex*, and *Notropis leedsi*. The sample was toxic to the Microtox® assay test organism, *Photobacterium phosphoreum*, producing an EC_{50} of 95.5% sample.

Chemical analyses were performed as part of the Compliance Sampling Toxics (CST) program. Trace amounts of possible petroleum hydrocarbons and numerous small, unidentified pesticide components were detected, as well as the metals, arsenic (8.6 µg/l), chromium (57 µg/l), lead (6 µg/l), and zinc (27 µg/l) (Table 4).

The cause of the observed toxicity in the Microtox® assay was not determined; however, the components detected by chemical analysis may have contributed to the observed toxicity.

Introduction

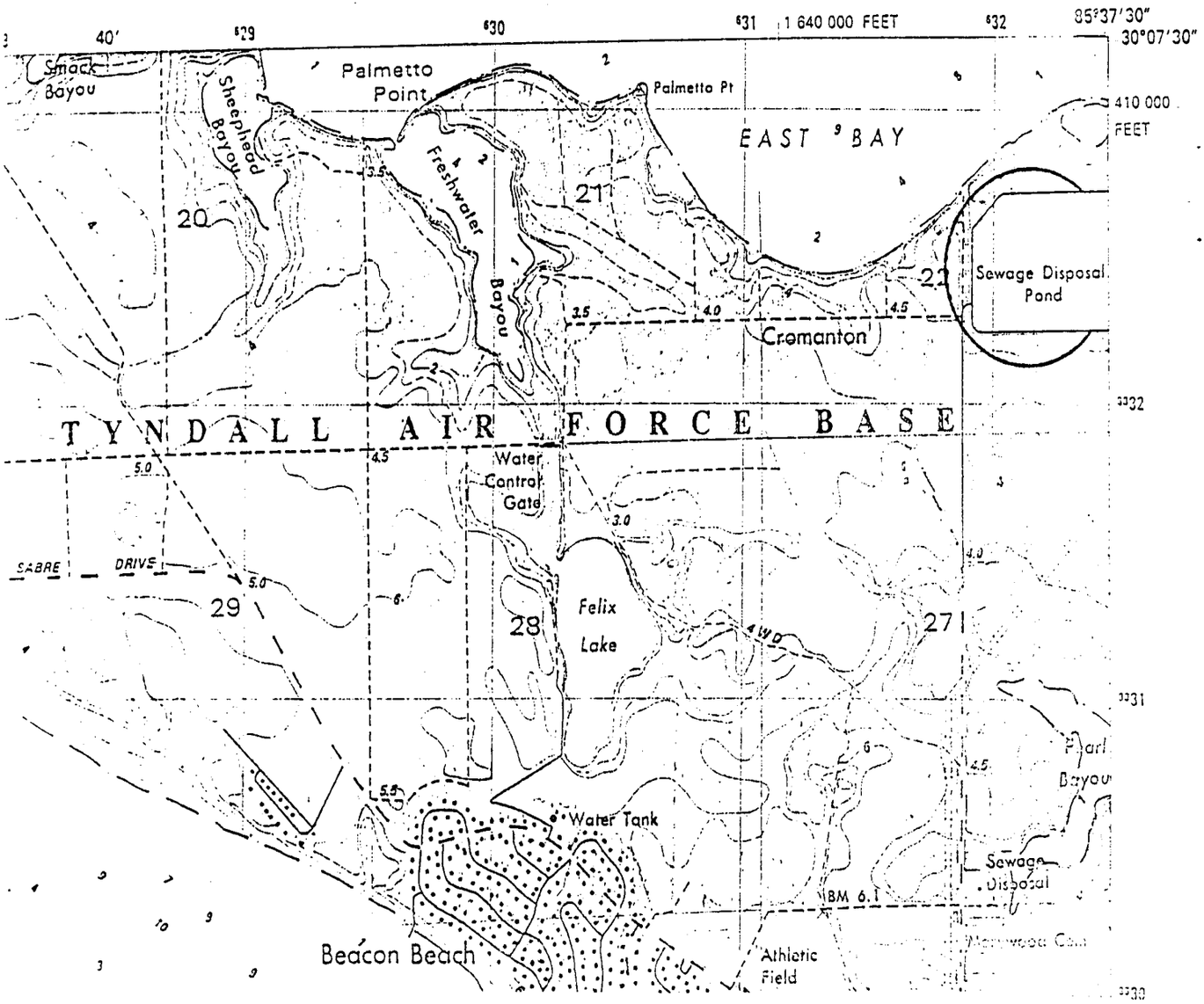
The Bay County Wastewater Treatment Plant final treatment process, a 59 acre aerated lagoon, is located on Tyndall Air Force Base, across St. Andrew Bay from the Southwest Forest/Arizona Chemical Company paper mill complex (Figure 1). The Bay County facility receives approximately 32 MGD of discharge from the paper mill complex for treatment. Pre-treatment at the complex includes evaporator waste condensers, primary clarification, and nutrient addition (Figure 2). Domestic discharges from at least three sources, the Millville STP, the Bay County System, and Tyndall AFB, may also be treated at the Bay County final aerated lagoon. Pre-treatment of these discharges may include degritting, primary treatment, and chlorination, prior to being pumped to the aerated lagoon (according to the NPDES file). After approximately 10 days retention in the aerated lagoon, the recovered water is discharged through a 1300 ft long outfall to St. Andrew Bay at a depth of approximately 15 feet. Additionally, this facility has a designated mixing zone in the bay (Stephen Baisden, personal communication) (FDER 1987).

The Biology Section had performed bioassays on samples from this facility in 1980, 1983, and 1986. In September, 1980, the tests using *Daphnia pulex* and *Mysidopsis bahia* indicated the sample was not acutely toxic. However, these tests were invalidated because the holding time of the samples was exceeded. No report was issued for the September, 1980 tests.

In July, 1983, a sample was acutely toxic to the test organism *M. bahia* (FDER 1983). In September, 1985, a sample was not acutely toxic to the test organisms *Notropis leedsi*, *D. pulex*, and *M. bahia*. In these tests, 15% of the *N. leedsi* died in the 100% sample test. No mortality occurred in the *M. bahia* test. The *D. pulex* test was invalidated due to excessive control mortality, but no mortality had occurred in the 100% sample at 24 hours (FDER 1985). Chemical analyses detected ammonia (maximum measured = 2.3 mg/l total and 0.01 mg/l unionized), several unidentified organic compounds, diethylphthalate (11 µg/l), lubricating oils, and the metals chromium (72 µg/l) and zinc (64 µg/l) (FDER 1985).

In December, 1986, the Biology Section performed bioassays on a sample from the facility using *D. pulex, N. leedsi, M. bahia*, and *Ceriodaphnia dubia* as the test organisms. The sample was not acutely toxic to the test organisms, *C. dubia, D. pulex,* and *N. leedsi*, but was sufficiently toxic to *M. bahia* to produce an LC_{50} of 118.6% sample. However, since this facility has a mixing zone, that LC_{50} was in compliance with Chapter 17-4 end-of-pipe toxicity standards (FDER 1987).

To again evaluate the existence and extent of the toxicity of this recovered water, the Biology Section performed three acute toxicity bioassays and one Microtox® assay on a sample from this facility on 7 to 9 June, 1988. 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.



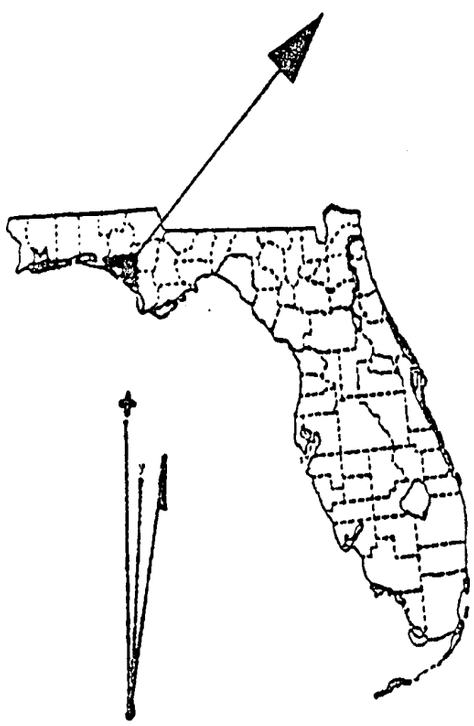


Figure 1. Portions of Bay County, Florida including Tyndall Air Force Base and the location of the Bay County WWTP.

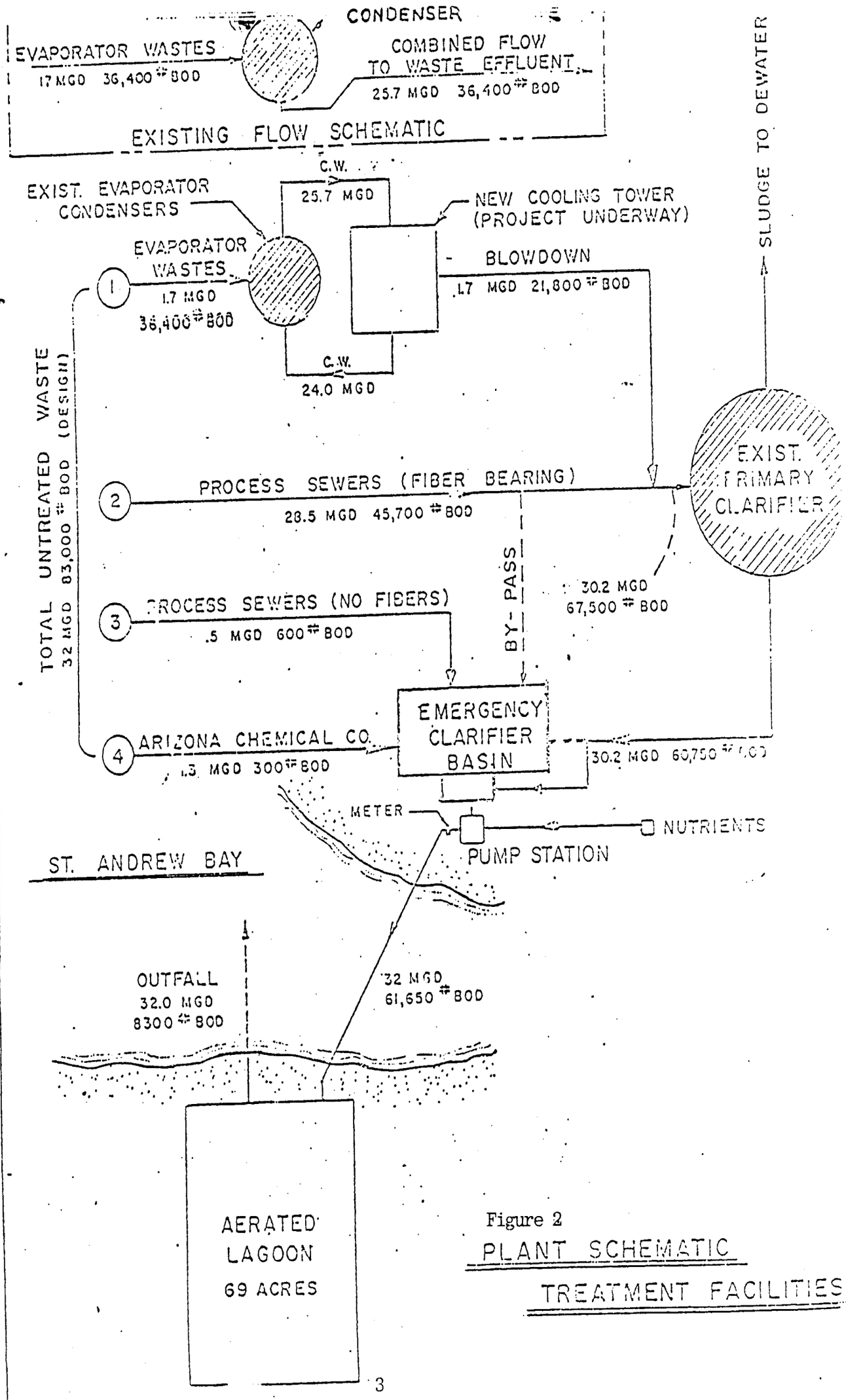


Figure 2

PLANT SCHEMATIC

TREATMENT FACILITIES

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 18 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

Three separate 48 hour acute toxicity bioassays and one Microtox® assay was performed on a sample from the Bay County WWTP. The tests were performed from 1000 hours on 7 June, 1988 to 1000 hours on 9 June, 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. Mark Clayton of the FDER Northwest District Branch office in Panama City, Bay County, Florida, 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.

Two replicates of the control and each test concentration were used in bioassays with the water fleas, *Ceriodaphnia dubia* and *Daphnia pulex*. Plastic vials of 30ml capacity (for C. dubia) 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

Three 48 hour acute toxicity bioassays were peformed on a sample from the aeration Lagoon Weir of the Bay County of the Bay County WWTP. The sample was not toxic to the bioassay test organisms, *Ceriodaphnia dubia, Daphnia pulex,* and *Notropis leedsi.* The sample was toxic to the Microtox® assay test organism, *Photobacterium phosphoreum,* producing on EC50 of 95.5% sample.

The total ammonia was 2.73 mg/l and the unionized ammonia was 0.03 mg/l. The sample was not chlorinated. Chemical analyses were performed as part of the Compliance Sampling Toxics (CST) program. Trace amounts of possible petroleum hydrocarbons and numerous small, unidentified pesticide components, as well as the metals, arsenic (8.6 µg/l), chromium (57 µg/l), lead (6 µg/l), and zinc (27 µg/l), were detected.

In conclusion, the sample from the Bay County WWTP was not acutely toxic to the bioassay organisms, *C. dubia, D. pulex,* and *N. leedsi*, but was acutely toxic to the Microtox® organism, *P. phosphoreum.* The cause of toxicity was not determined. The components detected by chemical analysis may have contributed to the observed toxicity..

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
30.0%	10.5 ml	4.5 ml	15 ml
60.0%	6.0 ml	9.0 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
30.0%	70.0 ml	30.0 ml	100 ml
60.0%	40.0 ml	60.0 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
30.0%	350 ml	150 ml	500 ml
60.0%	200 ml	300 ml	500 ml
100.0%	0 ml	500 ml	500 ml

Table 2. Data recorded during 48 hour static acute toxicity bioassay of the discharge from the Bay County WWTP, Panama City, Bay County, Florida, NPDES #FL0002631, on 7 to 9 June, 1988.

Facility:	Bay County WWTP			NPDES #:	FI#0002631	Facility Type:	Municipal STP
Location:	Panama City , Florida	County:	Bay County	Contact/District:	M. Clayton / NW		
				Test type:	Static Screening		
Sample Collection Date:	6/6/88	Time:	1435	# tests:	3	Receiving Water:	St. Andrews Bay
Test Beginning Date:	6/7/88	Time:	1000	Chlorination Type:	nonchlorinated		
Test Ending Date:	6/9/88	Time:	1000				

	Parameter	
Total Residual Cl2:	0.0	-
Method:	HACH	-
Salinity (ppt):	0	

Ammonia:	Control Water	100% Sample
Unionized NH3:	0.00	0.03
Total NH3:	0.00	2.73

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	3	9.2	-	9.0	8.0	-	7.8	-	-	-	-	-
Control	5	4	4	9.2	-	8.9	8.0	-	7.9	-	-	-	-	-
Control	5	4	3	9.3	-	9.0	8.0	-	7.9	-	-	-	-	-
Control	5	3	3	9.3	-	9.1	8.0	-	7.9	-	-	-	-	-
30%	5	5	4	9.1	-	9.0	7.7	-	7.9	-	-	-	-	-
30%	5	5	5	9.1	-	9.0	7.7	-	7.9	-	-	-	-	-
30%	5	5	5	9.1	-	9.1	7.7	-	7.9	-	-	-	-	-
30%	5	5	5	9.1	-	9.0	7.7	-	7.9	-	-	-	-	-
60%	5	5	5	9.0	-	8.9	7.5	-	7.9	-	-	-	-	-
60%	5	5	5	9.0	-	8.9	7.5	-	7.9	-	-	-	-	-
60%	5	5	5	9.0	-	9.0	7.5	-	7.9	-	-	-	-	-
60%	5	5	5	9.0	-	9.0	7.5	-	7.9	-	-	-	-	-
100%	5	5	5	8.9	-	8.9	7.4	-	8.0	-	-	-	-	-
100%	5	4	4	8.9	-	8.8	7.4	-	8.0	-	-	-	-	-
100%	5	5	5	8.9	-	8.9	7.4	-	7.9	-	-	-	-	-
100%	5	5	5	8.9	-	8.9	7.4	-	8.0	-	-	-	-	-

	Control Water	100% Sample
Alkalinity:	107	133
Hardness:	113	147

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	10	10	7	9.2	8.4	9.1	7.9	7.8	7.7	20.1	20.6	18.8	-	-
Control	10	10	10	9.3	8.5	9.1	7.9	7.9	7.8	-	-	-	-	-
30%	10	10	10	9.2	8.3	9.0	7.7	7.9	7.8	20.4	20.0	18.4	-	-
30%	10	10	10	9.2	8.3	9.0	7.7	7.9	7.8	-	-	-	-	-
60%	10	10	9	9.0	8.1	8.8	7.5	7.9	7.8	20.6	19.9	18.6	-	-
60%	10	10	10	9.0	8.1	8.9	7.5	7.9	7.8	-	-	-	-	-
100%	10	9	9	8.6	7.7	8.5	7.3	7.9	7.8	20.9	20.3	19.2	-	-
100%	10	10	9	8.6	7.6	8.4	7.3	7.9	7.8	-	-	-	-	-

	Control water	100% sample
Alkalinity:	107	133
Hardness:	113	147

Table 2. Continued.

Facility:	**Bay County WWTP**		NPDES #:	**Fl#0002631**	Facility Type: **Municipal STP**
Location:	Panama City , Florida	County: Bay County	Contact/District:	M. Clayton / NW	
			Test type:	Static Screening	
Sample Collection Date:	6/6/88	Time: 1435	# tests:	3	Receiving Water: St. Andrews Bay
Test Beginning Date:	6/7/88	Time: 1000	Chlorination Type:	**nonchlorinated**	
Test Ending Date:	6/9/88	Time: 1000			

	Parameter	
Total Residual Cl2:	0.0	-
Method:	HACH	-
Salinity (ppt):	0	

Ammonia:	Control Water	100% Sample
Unionized NH3:	0.00	0.03
Total NH3:	0.00	2.73

Organism: ***Notropis leedsi*** Age: 18 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	9.4	8.1	8.8	7.9	7.8	7.6	20.7	20.3	18.7	-	-
Control	5	5	5	9.4	8.1	8.8	7.9	7.8	7.6	-	-	-	240	232
30%	5	5	5	9.2	7.4	8.0	7.7	7.7	7.5	20.8	19.8	18.4	-	-
30%	5	5	5	9.2	7.4	8.0	7.7	7.7	7.5	-	-	-	780	710
60%	5	5	5	9.0	6.8	7.4	7.5	7.6	7.5	21.0	19.7	18.4	-	-
60%	5	5	5	9.0	6.8	7.2	7.5	7.6	7.5	-	-	-	1280	1190
100%	5	5	5	8.8	5.7	6.4	7.4	7.5	7.5	21.3	19.8	18.4	-	-
100%	5	5	5	8.8	5.8	6.4	7.4	7.5	7.5	-	-	-	1900	1800

	Control water	100% sample
Alkalinity:	120	133
Hardness:	125	147

Table 3. Microtox test results, Bay County WWTP, Bay County, NPDES #FL0002631, on 7 June, 1988.

Microtox Test — Culture Lot # M-606

% Sample	Final Reclaimed Water nonchlorinated 5-min	15-min
0	95	59
0	78	59
0	80	51
12.38	63	32
12.38	69	35
12.38	65	33
24.75	51	22
24.75	49	21
24.75	48	21
49.5	34	12
49.5	32	11
49.5	33	12
99	17	6
99	17	6
99	17	6

EC_{50} Nonchlorinated (15 minute, 15 °C) = 95.5% 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 4. Chemical components detected in the sample of discharge water from the Bay County WWTP, Panama City, Bay County, Florida, NPDES #FL0002631, on 6 June, 1988.

Organics

Acid Extractables	None Detected
Base Neutral Extractables	Trace amounts of possible petroleum hydrocarbons
Pesticide Extractables	Numerous small unidentified components

Metals

Arsenic	8.6 µg/l
Chromium	57 µg/l
Lead	6 µg/l
Zinc	27 µg/l

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.

Florida Department of Environmental Regulation. 1983. Bioassay of Bay County Wastewater Treatment Plant, Panama City Florida, NPDES #FL0002631. Biology Section. 11 p.

_____. 1985. Bioassays of Bay County Wastewater Treatment Plant, Panama City, Bay County, Florida, NPDES #FL0002631. Biology Section. 14 p.

_____. 1987. Bioassays of Bay County Wastewater Treatment Plant, Panama City, Bay County, Florida, NPDES #FL0002631. Biology Section. 13 p.

Bay County WWTP

Bay County

Outfall Number: 001

Permit Number:

Test Type: 7-day (Static) (Renewal) (Flow-through) (Screening) (Definitive) (Chronic)

Other: 3 1oz vials

Test Species: C. dubia neonate

6-6-88 1435
6-7-88 1000
6-9-88 1000

Concentration	Replicate	Number Initially Exposed	Number of Live Organisms 24	48	0	72	96	Dissolved Oxygen (mg/l) 0	24	48	72	96	Temperature °C 0	24	48	72	96	pH 0	24	48	72	96	Conductivity/Hardness (µmhos/cm) 0	24	48	72	96
Control	1/2	5/4	3/4	5/5				9.2/9.2		9.0/9.9			8.0/8.0		7.8/7.9													
Control	3/4	4/3	3/3	5/5				9.3/9.3		9.0/9.1			8.0/8.0		7.9/7.9													
30%	5	5	4	5				9.1		9.0			7.7		7.9													
30%	6	5	5	5				9.1		9.0			7.7		7.9													
30%	7	5	5	5				9.1		9.1			7.7		7.9													
30%	8	5	5	5				9.1		9.0			7.9		7.9													
60%	9	5	5	5				9.0		8.9			7.5		7.9													
60%	10	5	5	5				9.0		8.9			7.5		7.9													
60%	11	5	5	5				9.0		9.0			7.5		7.9													
60%	12	5	5	5				9.0		9.0			7.5		7.9													
100%	13	5	5	5				8.9		8.9			7.4		8.0			20.9										
100%	14	4	4	5				8.9		8.8			7.4		8.0													
100%	15	5	5	5				8.8		8.9			7.4		7.9													
100%	16	5	5	5				8.9		8.9			7.4		8.0													
Initial of Investigator		SHW	SHW	SHW				SHW		HF			RBF		HF													

Control Water: 107/11
100% Sample: 133/14
Other:
Measured by: SHW

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

Control Water: .1
100% Sample: .27
Other:

Hour

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

"0" slope -57.8

24 48 0

Signatures:

K M Lurding
Marshall Fischel
Russel Ingleberry
Steve Howe

Instrument Calibrations:

	pH Meter:	DO Meter:	S-C-T Meter:
0 Hours	7 at 7; 9 at 9	9.2 at 21.0°	
24 Hours	7 at 7; 9 at 9	8.8 at 22.0°	
48 Hours	7 at 7; 9 at 9	9.3 @ 19.0°C	1410, 21@3.0
72 Hours			
96 Hours			

Remarks: Conductivity Calibration
1310 at 1300
48 hr. 1280 @ 20°C (1273,0)

Total Residual Chlorine:

Field data: _____ Method: _____
Lab: 0.0 Method: HACH
Lab: _____ Method: _____

De-chlorinated:
Measurements by: _____

Reviewed by:

Date: 9/22/88

Industry/Toxicant: Bay County WWTP
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 6-6-88 Time 1435
Test Beginning: Date 6-7-88 Time 1000
Test Ending: Date 6-9-88 Time 1000
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	0	24	48	72	96
Control	—	10	10	7			9.2	8.4	9.1			7.9	7.8	7.7			20.1	20.6	18.8							
Control	25	10	10	10			9.3	8.5	9.1			7.9	7.9	7.8												
30%	56	10	10	10			9.2	8.3	9.0			7.7	7.9	7.8			20.4	20.0	18.4							
30%	7	10	10	10			9.2	8.3	9.0			7.7	7.9	7.8												
60%	38	10	10	9			9.0	8.1	8.8			7.5	7.9	7.8			20.6	19.9	18.6							
60%	65	10	10	10			9.0	8.1	8.9			7.5	7.9	7.8												
100%	70	10	9	9			8.6	7.7	8.5			7.3	7.9	7.8			20.9	20.3	19.2							
100%	13	10	10	9			8.6	7.6	8.4			9.3	7.9	7.8												
Initial of Investigator		RBF	MF	MF			SHW	SHW	MF			RBF	SHW	MF			RBF	MF	MF							

Alkalinity/Hardness (mg/l as $CaCO_3$)
Control Water: 107/113
100% Sample: ~~117/113~~ 133/147
Other:
Measured by: SHW

Ammonia unionized/total (mg/l as NH_3/N)
Hour 0
Control Water: 0.15
100% Sample: 2.73
Other:

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

Signatures:
R.B. Furday
Kenneth Sandell
Russell Frydenborg
Steven H. Woock

Instrument Calibrations:

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

Total Residual Chlorine:
Field data: ____ Method: ____
Lab: ____ Method: ____
Lab: ____ Method: ____
Dechlorinated:
Dechlorinated by:

Reviewed by:
Date: 9/26/88

Bay County WWTP

Test Number: 3 3

Test Type: (Acute) Chronic Effluent/Receiving Water/Conventional

Test Organism: N. Iglecta

Age/Life Stage: 18 days

Test Started: Date 6-6-88 Time 1435

Test Ended: Date 6-7-88 Time 1000

Test Duration: Date 6-7-88 Time 1000

Replicate	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
Control	16	5	5	5			9.4	8.1	8.8			7.9	7.8	7.6			20.7	20.3	18.7		
Control	131	5	5	5			9.4	8.1	8.8			7.9	7.8	7.6							
30%	7	5	5	5			9.2	7.4	8.0			7.7	7.7	7.5			20.8	19.8	18.4		
30%	5	5	5	5			9.2	7.4	8.0			7.7	7.7	7.5			KMW				
60%	113	5	5	5			9.0	6.8	7.4			7.5	7.6	7.5			20.0	19.7	18.4		
60%	46	5	5	5			9.0	6.8	7.2			7.5	7.6	7.5							
100%	43	5	5	5			8.8	5.7	6.4			7.4	7.5	7.5			21.3	19.8	18.4		
100%	54	5	5	5			8.8	5.8	6.4			7.4	7.5	7.5							
Initial of Investigator		KMW	MF	SHW			SHW	SHW	MF			RBF	SHW	MF			RBF	MF	MF		

Conductivity

Replicate	0	24	48	72	96
Control					
Control	240		232		
30%	780		710		
30%					
60%	1280		1190		
60%					
100%	1900		1800		
100%					

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

Control Water: 120 125 ~~107 113~~

100% Sample: 133/147

Other:

Measured by: SHW

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

Hour 0

Control Water: 0.0

100% Sample: 2.73

Other:

Hour ____

Control Water:

100% Sample:

Other:

Measured by: ____

Signatures:

KM Lurding

Marshall Faircloth

Russell Frydenborg

Steven Hibbs

Instrument Calibrations:

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

Total Residual Chlorine:

Field data: ____ Method: ____

Lab: ____ Method: ____

Lab: ____ Method: ____

De-chlorinated:

Measurements by: ____

Reviewed by: [signature]

Date: 9/26/88

Remarks: Conditioned water used for diluent -- well broken.

094

MICROTOX™ Data Reduction Sheet

SOURCE: Bay County STP

GRAPH CALC EC 50 (15 MIN, 15 °C) = 95.6 %SAMPLE mg/L

SAMPLE DATA:

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

DATE ASSAYED: 6-07 88 TIME: 9:00 Am

VISUAL COLOR: tannish 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: none

OPERATOR: Lesa Willis REAGENT VIAL LOT NO. M-606

REMARKS:

MICROTOX™ 93% TEST

SAMPLE CUVETTE	% SAMPLE	READINGS 5 MIN	READINGS 15 MIN	Γ5	Γ15	NOTES
A1	0	95	72	X	X	
B1		96	72			
C1		98	73			
A2	12.38	98	74	-.007	-.014	
B2		95	71			
C2		98	75			
A3	24.75	88	64	.086	.119	
B3		90	65			
C3		88	65			
A4	49.5	74	53	.262	.331	
B4		76	54			
C4		79	56			
A5	99	52	35	.877	1.087	
B5		49	33			
C5		53	36			
EC_{50} (MIN, °C)				107.99	95.49	

REVIEWED BY:

DATE: 9/26/88