DEPARTMENT OF ENVIRONMENTAL REGULATION

STATE OF FLORIDA

BIOASSAYS OF THE CECIL FIELD NAVAL AIR STATION

SEWAGE TREATMENT PLANT

DUVAL COUNTY, FLORIDA

NPDES #FL0020966

SAMPLED 1/26/87

MAY 7, 1987

Biology Section
Division of Environmental Programs

BIOASSAYS OF THE CECIL FIELD NAVAL AIR STATION

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Biology Section
Bureau of Laboratories and
Special Programs
May 7, 1987

EXECUTIVE SUMMARY

Cecil Field NAS Sewage Treatment Plant, Duval County, NPDES #FL0020966, 27 to 29 January, 1987.

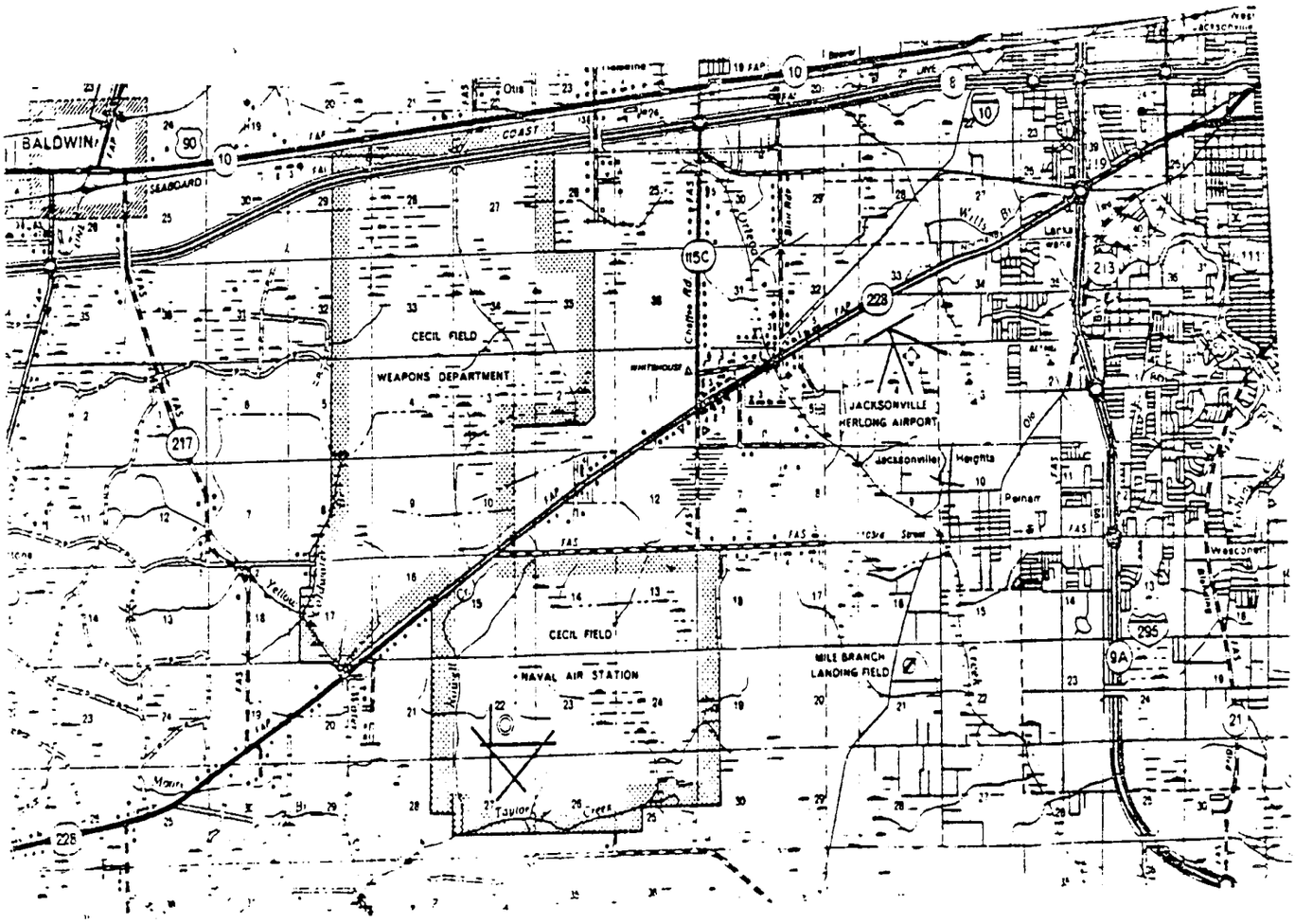
The sample of recovered water from this small government facility (< 1.0 MGD), which provides treatment for the Air Station personnel and its operations, was not acutely toxic to the test organisms, Ceriodaphnia dubia, Daphnia pulex, and Notropis leedsi. No significant mortality was noted during the 48 hour static tests. The chlorine (0.2 mg/l) and the ammonia (0.2 mg/l total and 0.001 mg/l unionized) detected in this sample were insufficient to have caused any toxicity.

INTRODUCTION

The Cecil Field Naval Air Station Sewage Treatment Plant serves the personnel of Cecil Field NAS, and has a design capacity of 0.82 MGD. Additionally, this plant provides secondary treatment for a variety of recycled waters generated as a result of the Naval Air Station's standard operating and maintenance procedures.

The plant discharges recovered water to the Caldwell Branch of Sal Taylor Creek (also known as Rowell Creek) which is a tributary of Yellow Water Creek (Figure 1). Yellow Water Creek eventually joins the North fork of Black Creek. Maintenance of good recovered water standards is especially important in this case because of the low flow and consequent minimal dilution capability of Yellow Water Creek.

In October, 1979, the Biology Section obtained samples from the Cecil Field Facility outfall #001 and performed static bioassays to determine the existence and extent of toxicity. The sample was not acutely toxic at that time (FDER 1979). Since it had been a number of years since this facility was tested, the Northeast District requested additional bioassays be performed to again evaluate the toxicity. Tests were performed from 12 to 14 February, 1986. Again, the sample was not acutely toxic (FDER 1986). Another series of bioassays was performed on samples from this facility by the Biology Section on 27 to 29 Janaury, 1987. The results of these latest tests are presented in this report. Those involved in performing these tests and in preparation of this report were: K. M. Lurding, M. L. Roll, and P. G. Weiland.



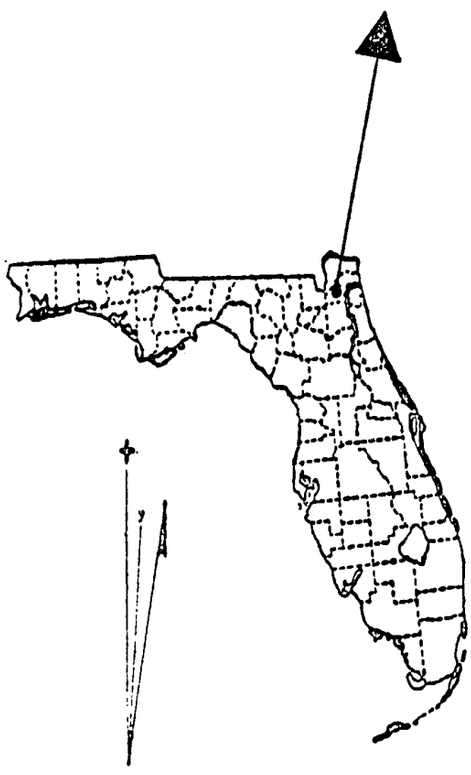


Figure 1. Portions of Duval County, Florida, including location of the Cecil Field Naval Air Station.

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 species recommended by Peltier and Weber (1985), the U.S. EPA (1978), 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.

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 March, 1979. 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 30 and 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 Tetra-Min® 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 Tetra Min®. The fish used in this test were approximately two weeks old at the time of the test.

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.

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 48 static acute toxicity bioassays were performed on a sample of recovered water from the Cecil Field STP outfall #001. The tests were performed from 1000 hours on 27 January to 1000 hours on 29 January, 1987.

The sample was collected by Mark Sittig of DER's Northeast District at 1115 hours on 26 January, 1987, using standard wastewater sampling procedures. The sample was 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 sample in the Tallahassee laboratory, the containers were placed in warm water and the temperature of the sample 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 recovered 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 sample. To accomplish this, the Biology Section performed concurrent bioassays on a sample of chlorinated water in which the chlorine had been neutralized. De-chlorination was accomplished by adding a calculated amount of 0.025N sodium thiosulfate. The total residual chlorine concentrations of the tested samples 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 recovered water.

Freshwater Tests

Freshwater for dilution of the samples 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 each concentration and a control, were used to test with the water fleas, Ceriodaphnia dubia and Daphnia pulex. Flint glass jars of either 60 ml (for C. dubia) or 250 ml (for D. pulex) capacity were used as test chambers, and either 50 ml or 200 ml 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, and loaded 10 per chamber, resulting in 20 per test concentration. Daphnia were siphoned into a large culture dish to isolate the proper size and age organisms (neonates). To accomplish this, the Daphnia were siphoned through a stainless steel screen. The trapped adults were then discarded. These neonates were then transferred by glass pipette, examined for injury, and were loaded 10 per chamber, resulting in 20 per test concentration. Water fleas were not fed during these tests.

Two replicates, of each concentration and a control, were used for the Notropis leedsi (bannerfin shiner) bioassays. The fish were transferred individually or by twos, into wide mouth quart jars containing 500 ml each of the appropriate test concentration, and were loaded 5 per chamber, resulting in 10 per concentration. Fish were not fed during these tests.

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% sample 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 daily 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% 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.

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

Ceriodaphnia dubia (water flea)

Concentration	Dilution Water	Sample	Total Volume
Control	50.0 ml	--------	50 ml
50.0%	25.0 ml	25.0 ml	50 ml
100.0%	-------	50.0 ml	50 ml

Daphnia pulex (water flea)

Concentration	Dilution Water	Sample	Total Volume
Control	200.0 ml	--------	200 ml
50.0%	100.0 ml	100.0 ml	200 ml
100.0%	--------	200.0 ml	200 ml

Notropis leedsi (bannerfin shiner)

Concentration	Dilution Water	Sample	Total Volume
Control	500 ml	--------	500 ml
50.0%	250 ml	250 ml	500 ml
100.0%	------	500 ml	500 ml

Table 2. Data recorded during 48 hour, *Ceriodaphnia dubia*, *Daphnia pulex*, and *Notropis leedsi*, static acute toxicity bioassays of Cecil Field NAS Sewage Treatment Plant, NPDES #FL0020966, on 27 to 29 January, 1987.

Chlorinated Sample[a]

Concentration or %	Number of Live Organisms			Dissolved Oxygen (mg/l)			pH			Temperature (°C)			Conductivity (umhos)	
	0	24	48	0	24	48	0	24	48	0	24	48	0	48
Ceriodaphnia dubia														
Control	10	10	10	8.0	8.2	7.3	8.0	7.9	7.7	20.6	20.5	20.7		
Control	10	10	10	8.0	8.2	7.3	8.0	7.9	7.7					
50.0%	10	10	10	8.3	8.4	7.9	8.0	8.0	7.8	20.3	20.2	20.5		
50.0%	10	10	10	8.3	8.4	7.9	8.0	8.0	7.8					
100.0%	10	10	10	8.5	8.3	7.9	7.2	8.0	7.8	20.2	20.2	20.5		
100.0%	10	10	10	8.5	8.3	7.9	7.2	8.0	7.8					
Daphnia pulex														
Control	10	10	10	7.9	8.0	7.5	8.0	7.9	7.7	21.0	20.4	20.4		
Control	10	10	10	7.9	8.0	7.5	8.0	7.9	7.7				215	215
50.0%	10	10	10	8.3	8.2	7.0	7.6	7.9	7.8	20.7	20.0	20.2		
50.0%	10	10	10	8.3	8.2	7.0	7.6	7.9	7.8				300	290
100.0%	10	10	10	8.7	8.4	6.5	7.1	7.9	7.8	20.5	20.0	20.2		
100.0%	10	10	10	8.7	8.4	7.1	7.1	7.9	7.8				370	370
Notropis leedsi														
Control	5	5	5	7.9	7.9	6.7	7.5	7.8	7.7	20.8	20.2	20.3		
Control	5	5	5	7.9	7.9	6.7	7.5	7.8	7.7				210	210
50.0%	5	5	5	8.3	8.3	7.3	7.4	7.8	7.7	20.6	19.8	20.1		
50.0%	5	5	5	8.3	8.3	7.3	7.4	7.8	7.7				295	290
100.0%	5	5	5	8.7	8.3	7.4	7.1	7.7	7.7	20.6	19.8	20.0		
100.0%	5	5	5	8.7	8.3	7.4	7.1	7.7	7.7				390	380

Control Water

NH_3(as NH_3)(mg/l): 0

Total ammonia (as N)(mg/l): 0

Total Alkalinity (mg/l $CaCO_3$): 120

Total Hardness (mg/l $CaCO_3$): 110

100% Sample

NH_3(as NH_3)(mg/l): .001

Total ammonia (as N)(mg/l): .2

Total Alkalinity (mg/l $CaCO_3$): 90

Total Hardness (mg/l $CaCO_3$): 100

[a] Total Chlorine Residual: 0.2 mg/l (HACH DPD Method).

Table 3. Data recorded during 48 hour, Ceriodaphnia dubia, Daphnia pulex, Notropis leedsi, static acute toxicity bioassays of Cecil Field NAS Sewage Treatment Plant, NPDES #FL0020066, on 27 to 29 January, 1987.

De-chlorinated Sample[a]

Concentration or %	Number of Live Organisms			Dissolved Oxygen (mg/l)			pH			Temperature (°C)			Conductivity (umhos)	
	0	24	48	0	24	48	0	24	48	0	24	48	0	48
Ceriodaphnia dubia														
Control	10	10	10	7.8	8.1	7.6	8.0	8.0	7.7	20.4	20.6	20.5		
Control	10	10	10	7.8	8.1	7.6	8.0	8.0	7.7					
50.0%	10	10	10	8.2	8.2	7.7	7.8	8.0	7.8	20.2	20.5	20.5		
50.0%	10	10	10	8.2	8.2	7.7	7.8	8.0	7.8					
100.0%	10	10	10	8.6	8.2	7.9	7.4	8.0	7.9	20.4	20.4	20.5		
100.0%	10	10	10	8.6	8.2	7.9	7.4	8.0	7.9					
Daphnia pulex														
Control	10	10	10	8.0	8.0	7.4	8.0	8.0	7.8	21.0	20.2	20.2		
Control	10	10	10	8.0	8.0	7.4	8.0	8.0	7.8				210	220
50.0%	10	10	10	8.5	7.9	7.4	7.6	7.9	7.8	20.8	20.0	20.1		
50.0%	10	10	10	8.5	7.9	7.4	7.6	7.9	7.8				290	300
100.0%	10	10	10	8.7	8.1	7.5	7.3	7.9	7.8	20.7	20.0	20.3		
100.0%	10	10	10	8.7	8.1	7.5	7.3	7.9	7.8				390	400
Notropis leedsi														
Control	5	5	5	7.9	7.9	6.7	7.5	7.8	7.7	20.8	20.2	20.3		
Control	5	5	5	7.9	7.9	6.7	7.5	7.8	7.7				210	210
50.0%	5	5	5	8.6	8.1	7.5	7.3	7.8	7.7	20.8	20.3	20.2		
50.0%	5	5	5	8.6	8.1	7.5	7.3	7.8	7.7				290	300
100.0%	5	5	5	9.0	8.0	7.7	7.2	7.8	7.7	20.7	20.3	20.4		
100.0%	5	5	5	9.0	8.0	7.7	7.2	7.8	7.7				390	390

Control Water	
NH_3(as NH_3)(mg/l) 0	Total Alkalinity (mg/l $CaOO_3$) 120
Total ammonia (as N)(mg/l) 0	Total Hardness (mg/l $CaOO_3$) 110

100% Sample	
NH_3(as NH_3)(mg/l) .001	Total Alkalinity (mg/l $CaOO_3$) 90
Total ammonia (as N)(mg/l) .2	Total Hardness (mg/l $CaOO_3$) 100

RESULTS AND DISCUSSION

The sample of recovered water from the Cecil Field Naval Air Station Sewage Treatment Plant was not acutely toxic to the test organisms, Ceriodaphnia dubia, Daphnia pulex, and Notropis leedsi. No significant mortality was noted during the 48 hours static tests (Tables 2 and 3). The residual chlorine (0.2 mg/l), and the ammonia (0.2 mg/l total and 0.001 mg/l unionized) detected in this sample were insufficient to have caused any toxicity.

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