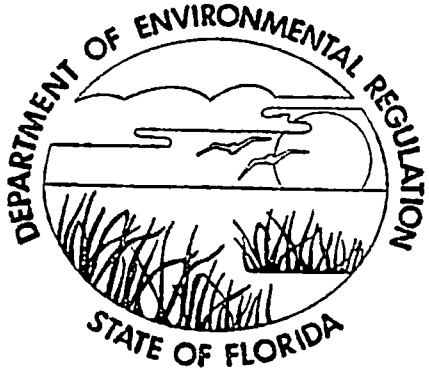


BIOASSAYS OF SOUTHERN WOOD PIEDMONT

BALDWIN, DUVAL COUNTY, FLORIDA

NPDES #FL0000221

MAY 18, 1987

Biology Section
Division of Environmental Programs

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EXECUTIVE SUMMARY

Southern Wood Piedmont, Baldwin, NPDES #FL0000221, on 11 to 13 February, 1987.

Three definitive static acute toxicity bioassays were performed on a sample of effluent collected from Southern Wood Piedmont's Outfall #002. Results indicated no toxicity to any of the test organisms, Ceriodaphnia dubia, Daphnia pulex, and Notropis leedsi. No mortality was observed in the 48 hour test duration.

Chemical analysis revealed the presence of unidentified pesticide extractables, as well as several metals in varying concentrations. This combination of compounds, or their concentrations, was apparently not lethal to the test organisms at this time.

INTRODUCTION

The Southern Wood Piedmont Company is a wood treating operation located in Baldwin, Duval County, Florida, approximately 10 miles west of Jacksonville (Figure 1). The plant pressure-treats a minimum of 50,000 cubic feet of wood per week with creosote, pentachlorophenol, and a solution of heavy metals to produce utility poles and crossties that are protected against weathering and decay. Although an elaborate treatment system with no discharge is used for process wastewater, freshly treated wood is temporarily stored for drying near the non-contact water effluent ditch (Figure 2). Any significant rainfall may cause contaminated runoff, containing the aforementioned treatment chemicals, to enter the effluent ditch prior to its discharge to Baldwin Bay Swamp, which eventually drains into the St. Mary's River.

Results of a difinitive 96 hour flow-through bioassay (FDER 1979) indicated that the effluent from the Southern Wood Piedmont Company was toxic to the organisms tested. Additional 48 hour static bioassays, however, performed in May, 1981, indicated that the effluent was not toxic to the organisms tested. But, it was suggested that the lack of toxicity was expected, due to a lack of rainfall at the facility at that time (FDER 1981). Conversely, subsequent static bioassays performed in July, 1982, indicated acute toxicity to Heterandria formosa and mortality of some Daphnia pulex. At the time of this sampling, the area had received recent heavy rainfall. Additionally, the effluent contained significant amounts of arsenic, copper, and zinc (FDER 1982).

To continue evaluating the toxicity of Southern Wood Piedmont's effluent, the Biology Section performed additional static tests on 11 to 13 February, 1987. Results are presented in this report. Those involved in performing the tests and in preparation of this report were: R. B. Frydenborg, K. M. Lurding, M. L. Roll, and P. G. Weiland.

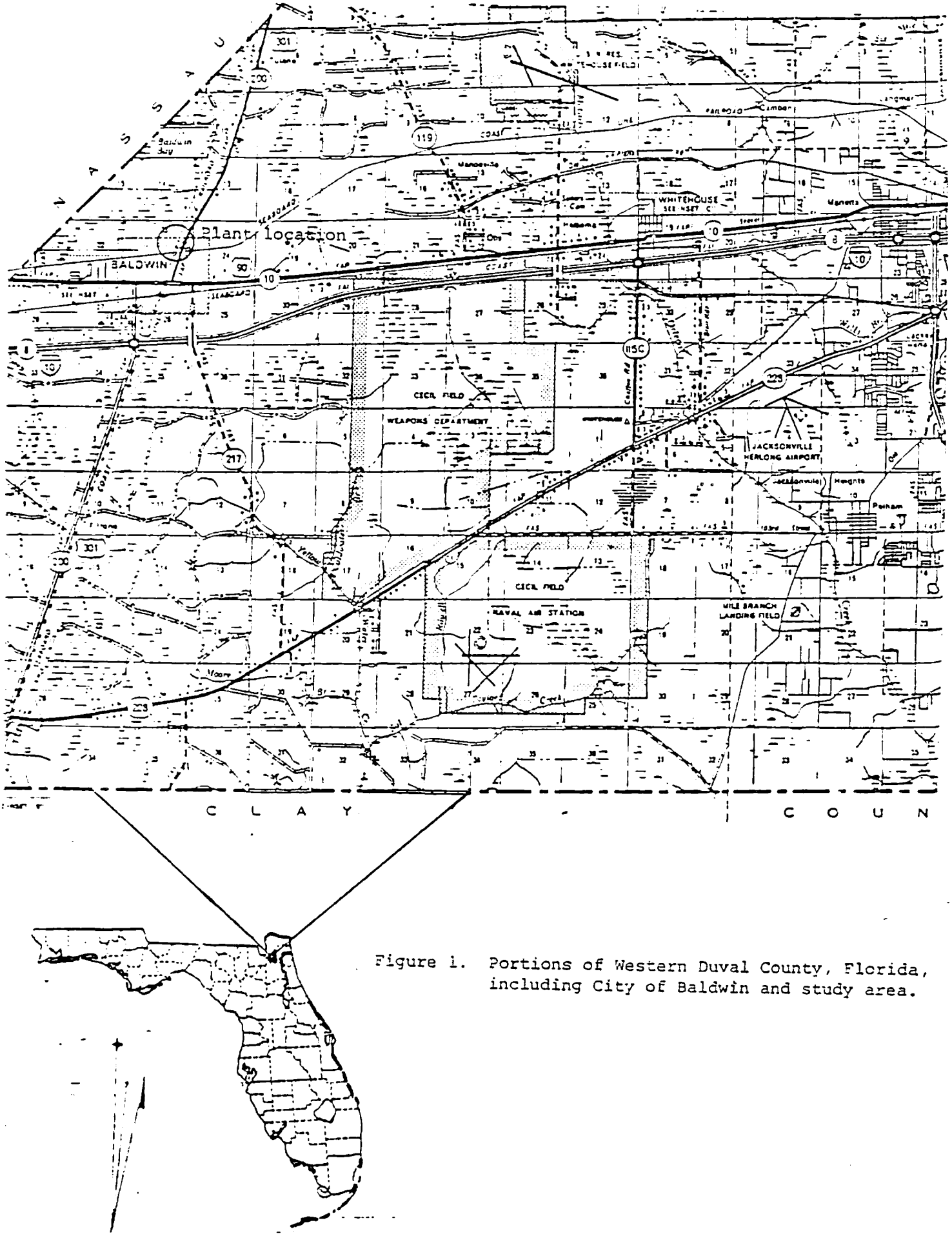
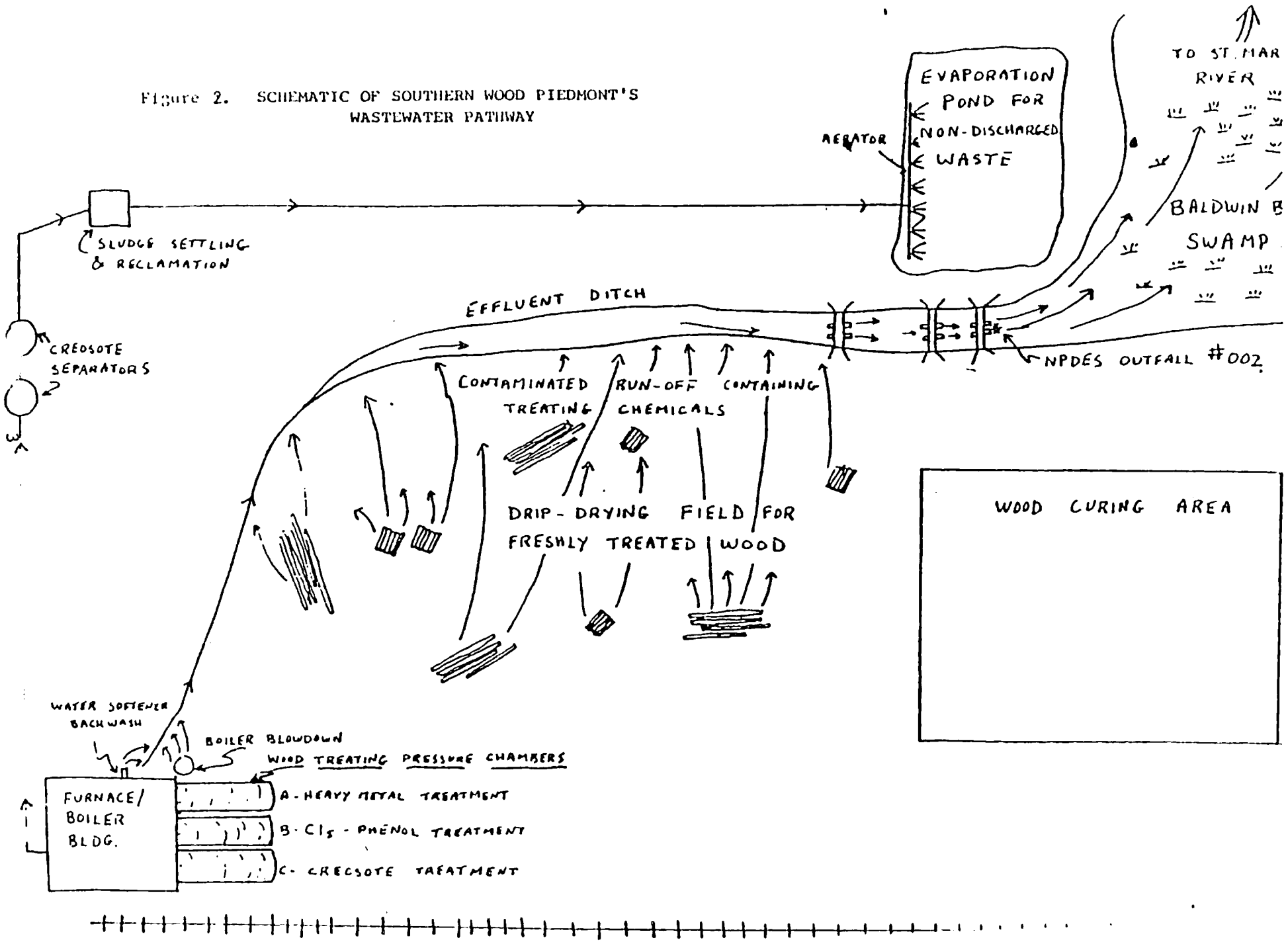


Figure 1. Portions of Western Duval County, Florida, including City of Baldwin and study area.

Figure 2. SCHEMATIC OF SOUTHERN WOOD PIEDMONT'S WASTEWATER PATHWAY



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 26 days 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

Three separate 48 hour definitive static acute toxicity bioassays were performed on a sample of Southern Wood Piedmont's discharge from Outfall #002. The tests were performed from 1030 hours on 11 February to 1030 hours on 13 February, 1987.

The samples were collected by Mr. Mark Sittig of DER's Northeast District Office at 0950 hours on 10 February, 1987, using standard wastewater sampling procedures. 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 iced and transported to the DER laboratory in Tallahassee for analyses within 25 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° $\pm$ 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.

Freshwater Tests

Freshwater for dilution of the effluent samples was obtained from the same DER deep well that is used to culture the test organisms. The concentration series selected for this study (Table 1) is an abbreviated version of that recommended by APHA (1980).

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) bioassay. 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 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% effluent 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% effluent 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, 3, and 4.

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

Ceriodaphnia dubia (water flea)

Concentration	Dilution Water	Effluent	Total Volume
Control	50.0 ml	--------	50 ml
10.0%	45.0 ml	5.0 ml	50 ml
32.0%	34.0 ml	16.0 ml	50 ml
56.0%	22.0 ml	28.0 ml	50 ml
100.0%	-------	50.0 ml	50 ml

Daphnia pulex (water flea)

Concentration	Dilution Water	Effluent	Total Volume
Control	200.0 ml	--------	200 ml
10.0%	180.0 ml	20.0 ml	200 ml
32.0%	136.0 ml	64.0 ml	200 ml
56.0%	88.0 ml	112.0 ml	200 ml
100.0%	--------	200.0 ml	200 ml

Notropis leedsi (bannerfin shiner)

Concentration	Dilution Water	Effluent	Total Volume
Control	500 ml	--------	500 ml
10.0%	450 ml	50 ml	500 ml
32.0%	340 ml	160 ml	500 ml
56.0%	220 ml	280 ml	500 ml
100.0%	------	500 ml	500 ml

Table 2. Data recorded during 48 hour, Ceriodaphnia dubia, static acute toxicity bioassay of Southern Wood Piedmont, NPDES #FL0000221, on 11 to 13 February, 1987.

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
Control	10	10	10	8.9	8.1	8.1	8.1	7.9	7.9	20.3	20.6	20.7		
Control	10	10	10	8.9	8.1	8.1	8.1	7.9	7.9					
10.0%	10	10	10	8.8	8.1	8.1	8.0	7.9	7.9	20.5	20.5	20.6		
10.0%	10	10	10	8.8	8.1	8.1	8.0	7.9	7.9					
32.0%	10	10	10	8.8	8.1	8.2	7.8	7.9	7.9	20.5	20.5	20.6		
32.0%	10	10	10	8.8	8.1	8.3	7.8	7.9	7.9					
56.0%	10	10	10	8.7	8.1	8.4	7.7	7.9	7.9	20.4	20.4	20.5		
56.0%	10	10	10	8.7	8.1	8.4	7.7	7.9	7.9					
100.0%	10	10	10	8.4	8.1	8.5	7.5	7.9	7.9	20.3	20.4	20.5		
100.0%	10	10	10	8.4	8.0	8.5	7.5	7.9	7.9					

Control Water

NH_3(as NH_3)(mg/l) 0

Total Alkalinity 120 (mg/l $CaOO_3$)

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

Total Hardness 115 (mg/l $CaOO_3$)

100% Effluent

NH_3(as NH_3)(mg/l) 0

Total Alkalinity 110 (mg/l $CaOO_3$)

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

Total Hardness 130 (mg/l $CaOO_3$)

Table 3. Data recorded during 48 hour, Daphnia pulex, static acute toxicity bioassay of Southern Wood Piedmont, NPDES #FL0000221, on 11 to 13, February, 1987.

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	
Control	10	10	10	9.0	8.1	8.2	8.0	7.9	7.9	20.9	20.5	20.5			Control Water
Control	10	10	10	9.0	8.1	8.2	8.0	7.9	7.9				210	210	NH_3(as NH_3)(mg/l) 0 / Total Alkalinity 120 (mg/l $CaCO_3$)
10.0%	10	10	10	9.0	8.2	8.3	8.0	7.9	7.9	21.0	20.4	20.3			
10.0%	10	10	10	9.0	8.2	8.3	8.0	7.9	7.9				210	210	
32.0%	10	10	10	8.9	8.2	8.3	7.8	7.9	7.9	21.0	20.2	20.2			Total ammonia (as N)(mg/l) 0 / Total Hardness 115 (mg/l $CaCO_3$)
32.0%	10	10	10	8.9	8.2	8.3	7.8	7.9	7.9				220	220	
56.0%	10	10	10	8.6	8.1	8.3	7.6	7.8	7.9	20.8	20.1	20.2			
56.0%	10	10	10	8.6	8.1	8.3	7.6	7.8	7.9				235	235	
100.0%	10	10	10	8.2	8.0	8.3	7.3	7.8	7.9	20.7	20.4	20.2			
100.0%	10	10	10	8.2	8.0	8.3	7.3	7.8	7.9				252	245	100% Effluent
															NH_3(as NH_3)(mg/l) 0 / Total Alkalinity 110 (mg/l $CaCO_3$)
															Total ammonia (as N)(mg/l) 0 / Total Hardness 130 (mg/l $CaCO_3$)

Table 4. Data recorded during 48 hour, Notropis leedsi, static acute toxicity bioassay of Southern Wood Piedmont, NPDES #FL0000221, on 11 to 13, February, 1987.

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	
Control	5	5	5	8.5	8.1	7.8	7.5	7.8	7.8	21.0	20.5	20.3			Control Water
Control	5	5	5	8.5	8.1	7.8	7.5	7.8	7.8				205	205	NH_3(as NH_3)(mg/l) 0 — Total Alkalinity 120 (mg/l $CaCO_3$)
10.0%	5	5	5	8.5	8.3	7.8	7.5	7.8	7.8	21.1	20.2	20.3			
10.0%	5	5	5	8.5	8.3	7.8	7.5	7.8	7.8				215	210	
32.0%	5	5	5	8.5	8.1	7.8	7.5	7.8	7.8	21.0	20.1	20.3			Total ammonia (as N)(mg/l) — Total Hardness 115 (mg/l $CaCO_3$)
32.0%	5	5	5	8.5	7.9	7.8	7.5	7.8	7.8				225	220	0
56.0%	5	5	5	8.3	7.7	7.7	7.4	7.7	7.8	20.8	20.1	20.2			
56.0%	5	5	5	8.3	7.7	7.7	7.4	7.7	7.8				240	230	
100.0%	5	5	5	8.2	7.7	7.6	7.4	7.7	7.8	20.7	20.4	20.3			
100.0%	5	5	5	8.2	7.7	7.6	7.4	7.7	7.8				255	250	100% Effluent
															NH_3(as NH_3)(mg/l) 0 — Total Alkalinity 110 (mg/l $CaCO_3$)
															Total ammonia (as N)(mg/l) — Total Hardness 130 (mg/l $CaCO_3$)
															0

RESULTS AND DISCUSSION

The effluent sample collected from Southern Wood Piedmont's Outfall #002 was not acutely toxic to any of the three test organisms, Ceriodaphnia dubia, Daphnia pulex, or Notropis leedsi. No mortality was noted during the 48 hour bioassay (Tables 2, 3, and 4).

Historically, bioassays performed on effluent samples from this facility have suggested an association of toxicity with heavy rainfall prior to sampling (FDER 1981, 1982). This was probably due to runoff from the area where freshly treated wood is temporarily stored for drying. Although at the time of this sampling it had not rained for 4 or 5 days, rainfall had previously been heavy, to the extent that backflow onto Southern Wood Piedmont's property had occurred (Mark Sittig, DER Northeast District Office, personal communication). The fact that this effluent sample was not toxic suggests either that most of the treatment chemicals were flushed out with the earlier heavy rainfall, or steps have been taken by the company to alleviate the problem.

The DER Chemistry Section analyzed samples of this effluent for certain organic compounds and metals (Table 5). No acid or base neutral extractables were detected, although the pesticide extractables analysis indicated several unidentified compounds. The metal analyses indicated chromium (86.0 ug/l), zinc (83 ug/l), arsenic (22.0 ug/l), lead 2.7 (ug/l), and trace amounts of copper. Apparently, these concentrations or particular combinations of chemical components were not toxic to the test organisms at this time.

Table 5. Results of Chemical Analyses Performed on Effluent from Southern Wood Piedmont, NPDES #FL0000221, collected 10 February, 1987.

Organics

Acid Extractables	N.D.
Base Neutral Extractables	N.D.
Pesticide Extractables	Several unidentified compounds

Metals

Antimony	N.D.
Arsenic	22.0 u/gl
Beryllium	N.D.
Cadmium	N.D.
Chromium	86.0 ug/l
Copper	< 25.0 ug/l[a]
Lead	2.7 ug/l
Mercury	[b]
Nickel	N.D.
Selenium	N.D.
Silver	N.D.
Thallium	N.D.
Zinc	83 ug/l

N.D. = None Detected

[a] Actual value is known to be less than value given.

[b] Results for mercury were invalidated due to contaminated field blank.

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