


# BIOASSAYS OF THE JACKSONVILLE U.S. NAVAL AIR STATION SEWAGE TREATMENT PLANT DISCHARGE COMBINED DOMESTIC AND INDUSTRIAL WASTEWATER JACKSONVILLE, DUVAL COUNTY, FLORIDA

NPDES #FL0000957

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MARCH 27, 1987

Biology Section
Division of Environmental Programs

# BIOASSAYS OF THE JACKSONVILLE U.S. NAVAL AIR STATION SEWAGE TREATMENT PLANT DISCHARGE COMBINED DOMESTIC AND INDUSTRIAL WASTEWATER JACKSONVILLE, DUVAL COUNTY, FLORIDA NPDES #FL0000957

Biology Section
Bureau of Laboratories and
Special Programs
March 27, 1987

## EXECUTIVE SUMMARY

Jacksonville NAS Sewage Treatment Plant Combined Domestic and Industrial Discharge, NPDES #FL0000957, on 13 to 15 January, 1987.

The final chlorinated sample from this U.S. government facility proved to be acutely toxic to the test organisms, Ceriodaphnia dubia, Daphnia pulex, and Notropis leedsi, producing 24 hour $LC_{50}$'s of < 10%, < 10%, and 17.9%, respectively. This toxicity can be mostly attributed to the total residual chlorine (1.0 mg/l HACH DPD Method) measured in this sample. This same sample after de-chlorination was not toxic to the same test organisms.

The ammonia detected in these samples (maximum = 0.08 mg/l unionized) was insufficient to have contributed to the toxicity. Chemical analyses indicated the presence of endosulfan, unidentified organic compounds, and the metals cadmium, chromium, silver, and zinc in the sample. These chemical components probably contributed to the toxicity noted.

# INTRODUCTION

The Jacksonville U.S. Naval Air Station is a U.S. Government facility located in Jacksonville, Duval County, Florida (Fig. 1). The associated sewage treatment plant treats waste from the air station, including domestic waste, water from industrial cooling systems, boiler wash, vehicle and equipment cleaning operations, and other miscellaneous sources. The plant has a design capacity of 3.0 MGD and is currently operating near this level, 2.6 MGD. Discharge is to the St. Johns River (NPDES Outfall #001).

The Biology Section had previously performed bioassays on pre-chlorinated, chlorinated, and de-chlorinated samples from this facility, in May, 1981. The results of those tests indicated that the pre-chlorinated and de-chlorinated samples were not toxic to Daphnia pulex, while the chlorinated sample was extremely toxic to Daphnia, with 24 and 48 hours $LC_{50}$ values of < 5.6% (FDER 1981). The toxicity was attributed to the presence of total residual chlorine.

To again evaluate the existence and extent of toxicity of this discharge, the Biology Section performed additional static acute toxicity bioasays on a grab sample, on 3 to 5 August, 1982. The results of those tests indicated that the sample was not toxic. Only a small residual chlorine (< 0.05 mg/1) was detected at that time (FDER 1982).

Additional bioassays were performed in August, 1983, on pre-, post-, and de-chlorinated samples from this facility. Once again, the chlorinated sample was extremely toxic to D. pulex ($LC_{50}$ = < 5.6%). Mortality was also noted in the pre- and de-chlorinated samples (FDER 1983).

In January, 1986, DER's Northeast District requested additional bioassays to further evaluate the Jacksonville NAS facility. Grab samples of recovered water from the sewage treatment system and from upstream of the industrial wastewater system were collected. The results of those tests indicated once again that the chlorinated sample was acutely toxic ($LC_{50}$'s approximately 10%) but that the de-chlorinated sample was not toxic. Some mortality was noted in the industrial sample, but an $LC_{50}$ could not be calculated.

In the latest series of tests of this facility, grab samples of final chlorinated recovered water, consisting of combined waters from the domestic waste and industrial treatment sections, were collected for bioassay analyses. The results of this latest series of bioassays are presented in this report. Those involved in performing the tests and in preparation of this report were: M. L. Roll and P. G. Weiland.

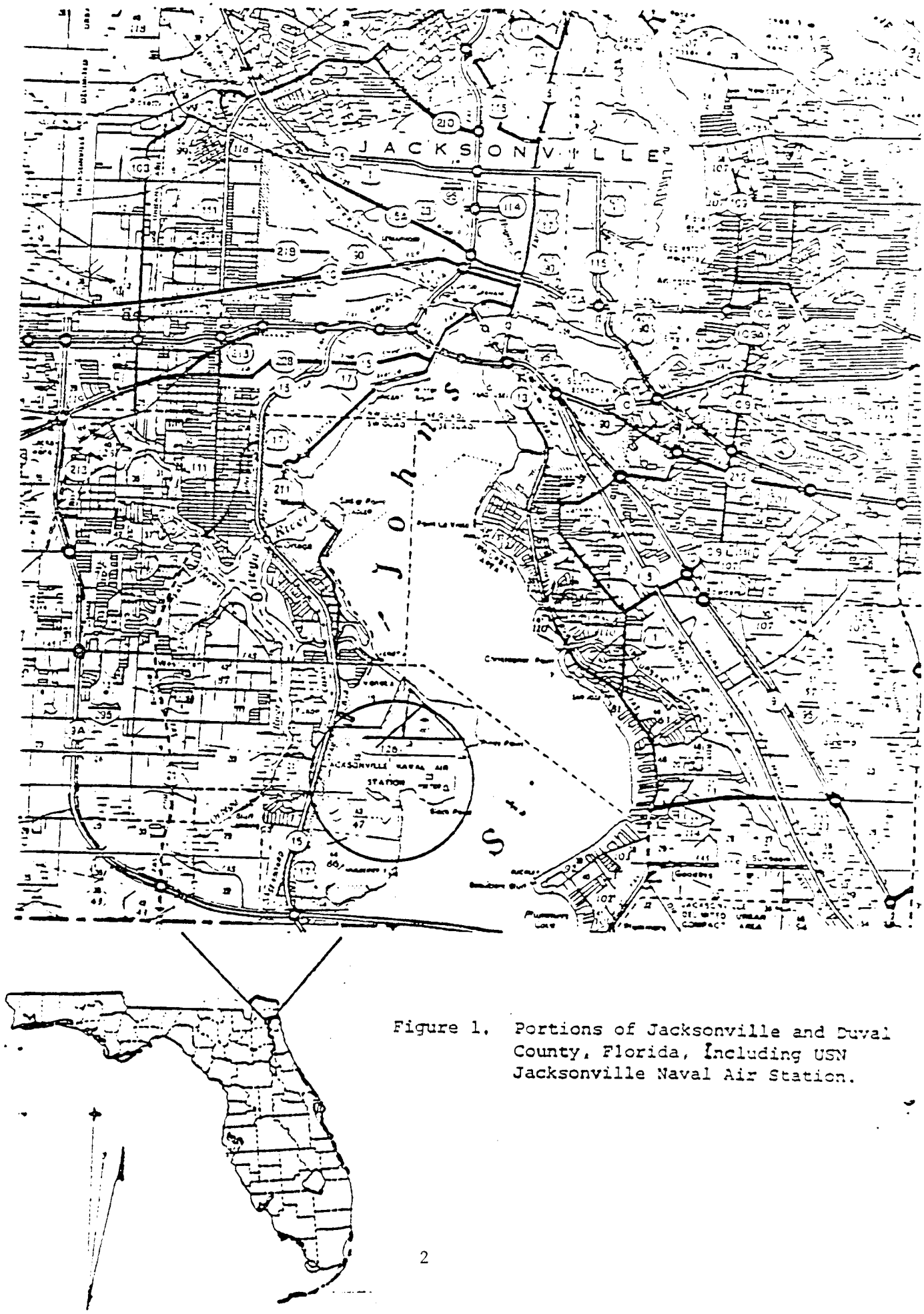



Figure 1. Portions of Jacksonville and Duval County, Florida, Including USN Jacksonville Naval Air Station.

## 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 a 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 32 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

Six separate 48 our static acute toxicity bioassays were performed on post-chlorinated and de-chlorinated samples of recovered water from the Jacksonville NAS facility. The tests were performed from 1000 hours on 13 January to 1000 hours on 15 January, 1987.

The samples were collected by Mr. Mark Sittig of DER's Northeast District at 1050 hours on 12 January, 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 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° $\pm$ 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.

### 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 water. 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 of 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 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 (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) 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% 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

concentrations 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 through 7.

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 |
| 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 | Sample | 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 | Sample | 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 Jacksonville NAS Sewage Treatment Plant, NPDES #FL0000957, on 13 to 15 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 | |
| Control | 10 | 10 | 10 | 8.4 | 8.1 | 8.2 | 8.0 | 7.9 | 7.9 | 20.3 | 19.9 | 21.0 | | | Control Water |
| Control | 10 | 10 | 8 | 8.4 | 8.1 | 8.2 | 8.0 | 7.9 | 7.9 | | | | | | $NH_3$(as $NH_3$)(mg/l) 0 — Total Alkalinity 119 (mg/l $CaOO_3$) |
| 10.0% | 10 | 1 | 0 | 8.5 | 8.5 | 8.2 | 8.0 | 7.9 | 7.9 | 20.4 | 19.7 | 21.0 | | | |
| 10.0% | 10 | 1 | 0 | 8.5 | 8.5 | 8.2 | 8.0 | 7.9 | 7.9 | | | | | | Total ammonia (as N)(mg/l) 0 — Total Hardness 113 (mg/l $CaOO_3$) |
| 32.0% | 10 | 0 | 0 | 8.1 | 8.7 | -b | 7.8 | 8.0 | -b | 20.4 | 19.6 | -b | | | |
| 32.0% | 10 | 0 | 0 | 8.1 | 8.7 | - | 7.8 | 8.0 | - | | | | | | |
| 56.0% | 10 | 0 | 0 | 8.7 | 8.7 | - | 7.7 | 8.1 | - | 20.6 | 19.5 | - | | | |
| 56.0% | 10 | 0 | 0 | 8.7 | 8.7 | - | 7.7 | 8.1 | - | | | | | | |
| 100.0% | 10 | 0 | 0 | 8.9 | 8.8 | - | 7.5 | 8.1 | - | 20.8 | 19.6 | - | | | |
| 100.0% | 10 | 0 | 0 | 8.9 | 8.8 | - | 7.5 | 8.1 | - | | | | | | 100% Effluent |
| | | | | | | | | | | | | | | | $NH_3$(as $NH_3$)(mg/l) 0.08 — Total Alkalinity 115 (mg/l $CaOO_3$) |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | Total ammonia (as N)(mg/l) 5.0 — Total Hardness 209 (mg/l $CaOO_3$) |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | |

[a] Total Residual Chlorine: 1.0 mg/l (HACH DPD Method), 0.71 mg/l (Iodometric Method I).

[b] Remaining parameters not recorded since all organisms were dead by 24 hours.

Table. 3. Data recorded during 48 hour, Daphnia pulex, static acute toxicity bioassay of Jacksonville NAS Sewage Treatment Plant, NPDES #FL0000957, on 13 to 15 January, 1987.

Chlorinated Sample[a]

| Concentration or % | Number of Live Organisms 0 | 24 | 48[b] | Dissolved Oxygen (mg/l) 0 | 24 | 48 | pH 0 | 24 | 48 | Temperature (°C) 0 | 24 | 48 | Conductivity (umhos) 0 | 48 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Control | 10 | 10 | | 8.3 | 8.4 | | 8.0 | 8.0 | | 20.5 | 19.5 | | | |
| Control | 10 | 10 | | 8.3 | 8.4 | | 8.0 | 8.0 | | | | | 212 | |
| 10.0% | 10 | 5 | | 8.4 | 8.4 | | 7.9 | 8.0 | | 20.5 | 19.3 | | | |
| 10.0% | 10 | 2 | | 8.4 | 8.4 | | 7.9 | 8.0 | | | | | 249 | |
| 32.0% | 10 | 0 | | 8.5 | 8.7 | | 7.8 | 8.0 | | 20.6 | 19.2 | | | |
| 32.0% | 10 | 0 | | 8.5 | 8.7 | | 7.8 | 8.0 | | | | | 312 | |
| 56.0% | 10 | 0 | | 8.7 | 8.8 | | 7.6 | 8.0 | | 20.7 | 19.1 | | | |
| 56.0% | 10 | 0 | | 8.7 | 8.8 | | 7.6 | 8.0 | | | | | 388 | |
| 100.0% | 10 | 0 | | 9.0 | 8.9 | | 7.5 | 8.0 | | 21.0 | 19.2 | | | |
| 100.0% | 10 | 0 | | 9.0 | 8.9 | | 7.5 | 8.0 | | | | | 600 | |

Control Water

$NH_3$(as $NH_3$)(mg/l) . 0

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

Total Alkalinity 119 (mg/l $CaOO_3$)

Total Hardness 113 (mg/l $CaOO_3$)

100% Effluent

$NH_3$(as $NH_3$)(mg/l) 0.08

Total ammonia (as N)(mg/l) 5.0

Total Alkalinity 115 (mg/l $CaOO_3$)

Total Hardness 209 (mg/l $CaOO_3$)

[a] Total Residual Chlorine: 1.0 mg/l (HACH DPD Method), 0.71 mg/l (Iodometric Method I).

[b] Test invalidated at 48 hours due to excessive control mortality (> 10%). Data not reported.

Table 4. Data recorded during 48 hour, Notropis leedsi, static acute toxicity bioassay of Jacksonville NAS Sewage Treatment Plant, NPDES #FL0000957, on 13 to 15 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 | |
| Control | 5 | 5 | 5 | 8.2 | 7.3 | 7.2 | 7.6 | 7.8 | 7.8 | 21.2 | 19.5 | 20.4 | | | Control Water |
| Control | 5 | 5 | 5 | 8.2 | 7.3 | 7.2 | 7.6 | 7.8 | 7.8 | | | | 212 | 210 | $NH_3$(as $NH_3$)(mg/l) 0 — Total Alkalinity 119 (mg/l $CaOO_3$) |
| 10.0% | 5 | 5 | 5 | 8.3 | 7.5 | 7.3 | 7.6 | 7.7 | 7.8 | 21.2 | 19.2 | 20.4 | | | |
| 10.0% | 5 | 5 | 5 | 8.3 | 7.5 | 7.0 | 7.6 | 7.7 | 7.8 | | | | 248 | 240 | |
| 32.0% | 5 | 0 | 0 | 8.5 | 8.3 | -b | 7.5 | 7.7 | -b | 21.1 | 19.0 | -b | | | Total ammonia (as N)(mg/l) — Total Hardness 113 (mg/l $CaOO_3$) |
| 32.0% | 5 | 0 | 0 | 8.5 | 8.3 | - | 7.5 | 7.7 | - | | | | 315 | -b | 0 |
| 56.0% | 5 | 0 | 0 | 8.8 | 8.4 | - | 7.5 | 7.7 | - | 21.0 | 18.8 | - | | | |
| 56.0% | 5 | 0 | 0 | 8.8 | 8.4 | - | 7.5 | 7.7 | - | | | | 392 | - | |
| 100.0% | 5 | 0 | 0 | 9.0 | 8.4 | - | 7.4 | 7.7 | - | 21.2 | 18.9 | - | | | |
| 100.0% | 5 | 0 | 0 | 9.0 | 8.4 | - | 7.4 | 7.7 | - | | | | 600 | - | 100% Effluent |
| | | | | | | | | | | | | | | | $NH_3$(as $NH_3$)(mg/l) 0.07 — Total Alkalinity 115 (mg/l $CaOO_3$) |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | Total ammonia (as N)(mg/l) 5.0 — Total Hardness 209 (mg/l $CaOO_3$) |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | |

[a] Total Residual Chlorine: 1.0 mg/l (HACH DPD Method), 0.71 mg/l (Iodometric Method I).

[b] Remaining parameters not recorded since all organisms were dead by 24 hours.

Table 5. Data recorded during 48 hour, Ceriodaphnia dubia, static acute toxicity bioassay of Jacksonville NAS Sewage Treatment Plant, NPDES #FL0000957, on 13 to 15 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 | |
| Control | 10 | 10 | 8 | 8.3 | 8.2 | 8.0 | 7.9 | 7.9 | 7.9 | 20.7 | 19.3 | 20.8 | | | Control Water |
| Control | 10 | 10 | 10 | 8.3 | 8.2 | 8.0 | 7.9 | 7.9 | 7.9 | | | | | | $NH_3$(as $NH_3$)(mg/l) 0 — Total Alkalinity 119 (mg/l $CaOO_3$) |
| 10.0% | 10 | 10 | 10 | 8.4 | 8.2 | 8.1 | 7.9 | 8.0 | 7.9 | 20.5 | 19.5 | 20.7 | | | |
| 10.0% | 10 | 10 | 8 | 8.4 | 8.2 | 8.1 | 7.9 | 8.0 | 7.9 | | | | | | Total ammonia (as N)(mg/l) 0 — Total Hardness 113 (mg/l $CaOO_3$) |
| 32.0% | 10 | 10 | 10 | 8.6 | 8.2 | 8.4 | 7.8 | 7.9 | 7.9 | 20.5 | 19.5 | 20.5 | | | |
| 32.0% | 10 | 10 | 9 | 8.6 | 8.2 | 8.4 | 7.8 | 7.9 | 7.9 | | | | | | |
| 56.0% | 10 | 10 | 10 | 8.8 | 8.1 | 8.3 | 7.7 | 7.9 | 7.9 | 20.5 | 19.5 | 20.5 | | | |
| 56.0% | 10 | 10 | 10 | 8.8 | 8.1 | 8.3 | 7.7 | 7.9 | 7.9 | | | | | | |
| 100.0% | 10 | 10 | 10 | 9.0 | 8.4 | 8.2 | 7.5 | 8.0 | 7.9 | 20.8 | 19.5 | 20.5 | | | |
| 100.0% | 10 | 10 | 10 | 9.0 | 8.4 | 8.2 | 7.5 | 8.0 | 7.9 | | | | | | 100% Effluent |
| | | | | | | | | | | | | | | | $NH_3$(as $NH_3$)(mg/l) — Total Alkalinity (mg/l $CaOO_3$) |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | Total ammonia (as N)(mg/l) — Total Hardness (mg/l $CaOO_3$) |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | |

[a] Total Residual Chlorine: None Detected (HACH DPD Method).

Table 6. Data recorded during 48 hour, Daphnia pulex, static acute toxicity bioassay of Jacksonville NAS Sewage Treatment Plant, NPDES #FL0000957, on 13 to 15 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 | | |
| Control | 10 | 10 | 10 | 8.3 | 8.1 | 8.2 | 8.0 | 8.0 | 7.9 | 20.7 | 19.4 | 20.5 | | | Control Water | |
| Control | 10 | 10 | 10 | 8.3 | 8.1 | 8.2 | 8.0 | 8.0 | 7.9 | | | | 218 | 230 | $NH_3$(as $NH_3$)(mg/l) | Total Alkalinity 119 (mg/l $CaOO_3$) |
| 10.0% | 10 | 10 | 10 | 8.4 | 8.2 | 8.3 | 7.8 | 8.0 | 7.9 | 20.5 | 19.2 | 20.2 | | | 0 | |
| 10.0% | 10 | 10 | 10 | 8.4 | 8.2 | 8.3 | 7.8 | 8.0 | 7.9 | | | | 255 | 260 | | |
| 32.0% | 10 | 10 | 10 | 8.6 | 8.2 | 8.3 | 7.8 | 8.0 | 7.9 | 20.8 | 19.1 | 20.1 | | | Total ammonia (as N)(mg/l) | Total Hardness 113 (mg/l $CaOO_3$) |
| 32.0% | 10 | 10 | 10 | 8.6 | 8.2 | 8.3 | 7.8 | 7.9 | 7.9 | | | | 325 | 330 | 0 | |
| 56.0% | 10 | 10 | 10 | 8.8 | 8.0 | 8.2 | 7.7 | 7.9 | 7.9 | 20.9 | 19.1 | 20.2 | | | | |
| 56.0% | 10 | 10 | 10 | 8.8 | 8.0 | 8.2 | 7.7 | 7.9 | 7.9 | | | | 400 | 400 | | |
| 100.0% | 10 | 10 | 10 | 9.0 | 8.5 | 7.9 | 7.5 | 7.9 | 7.9 | 21.1 | 19.3 | 20.3 | | | | |
| 100.0% | 10 | 10 | 10 | 9.0 | 8.5 | 7.9 | 7.5 | 7.9 | 7.9 | | | | 600 | 600 | 100% Effluent | |
| | | | | | | | | | | | | | | | $NH_3$(as $NH_3$)(mg/l) | Total Alkalinity 119 (mg/l $CaOO_3$) |
| | | | | | | | | | | | | | | | 0.08 | |
| | | | | | | | | | | | | | | | Total ammonia (as N)(mg/l) | Total Hardness 205 (mg/l $CaOO_3$) |
| | | | | | | | | | | | | | | | 5.0 | |

[a] Total Residual Chlorine: None Detected (HACH DPD Method).

Table 7. Data recorded during 48 hour, Notropis leedsi, static acute toxicity bioassay of Jacksonville NAS Sewage Treatment Plant, NPDES #FL0000957, on 13 to 15 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 | |
| Control | 5 | 5 | 5 | 8.2 | 7.3 | 7.2 | 7.6 | 7.8 | 7.8 | 21.2 | 19.5 | 20.4 | | | Control Water |
| Control | 5 | 5 | 5 | 8.2 | 7.3 | 7.2 | 7.6 | 7.8 | 7.8 | | | | 212 | 210 | $NH_3$(as $NH_3$)(mg/l) 0 / Total Alkalinity 119 (mg/l $CaOO_3$) |
| 10.0% | 5 | 5 | 5 | 8.4 | 7.8 | 7.7 | 7.5 | 7.8 | 7.7 | 21.0 | 19.1 | 20.3 | | | |
| 10.0% | 5 | 5 | 5 | 8.4 | 7.8 | 7.7 | 7.5 | 7.8 | 7.7 | | | | 242 | 240 | |
| 32.0% | 5 | 5 | 5 | 8.5 | 7.6 | 7.5 | 7.5 | 7.7 | 7.7 | 21.1 | 19.0 | 20.2 | | | Total ammonia (as N)(mg/l) 0 / Total Hardness 113 (mg/l $CaOO_3$) |
| 32.0% | 5 | 5 | 5 | 8.5 | 7.6 | 7.5 | 7.5 | 7.7 | 7.7 | | | | 318 | 310 | |
| 56.0% | 5 | 5 | 5 | 8.8 | 7.5 | 7.6 | 7.5 | 7.7 | 7.7 | 21.3 | 18.9 | 20.1 | | | |
| 56.0% | 5 | 5 | 5 | 8.8 | 7.5 | 7.6 | 7.5 | 7.7 | 7.7 | | | | 385 | 375 | |
| 100.0% | 5 | 5 | 5 | 9.0 | 7.5 | 7.3 | 7.4 | 7.7 | 7.7 | 21.3 | 19.1 | 20.1 | | | |
| 100.0% | 5 | 5 | 5 | 9.0 | 7.5 | 7.3 | 7.4 | 7.7 | 7.7 | | | | 600 | 600 | 100% Effluent |
| | | | | | | | | | | | | | | | $NH_3$(as $NH_3$)(mg/l) 0.07 / Total Alkalinity 119 (mg/l $CaOO_3$) |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | Total ammonia (as N)(mg/l) 5.0 / Total Hardness 205 (mg/l $CaOO_3$) |
| | | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | |

[a] Total Residual Chlorine: None Detected (HACH DPD Method).

Table 8. $LC_{50}$ Data for Bioassays of the Jacksonville NAS STP, NPDES #FL0000957, on 13 to 15 January, 1987.

| Organism | Method of Data Analysis | 24 Hour $LC_{50}$ and 95% Confidence Intervals | 48 Hour $LC_{50}$ and 95% Confidence Intervals |
|---|---|---|---|
| Chlorinated Sample | | | |
| Ceriodaphnia dubia | Binomial<br>Moving Average<br>Probit | < 10.0%, a | < 10.0%, a |
| Daphnia pulex | Binomial<br>Moving Average<br>Probit | < 10.0%, a | b |
| Notropis leedsi | Binomial<br>Moving Average<br>Probit | 17.9% (10 and 32)<br>c<br>c | 17.9% (10 and 32)<br>c<br>c |
| De-chlorinated Sample | | | |
| C. dubia | Binomial<br>Moving Average<br>Probit | No Mortality | Random Mortality |
| D. pulex | Binomial<br>Moving Average<br>Probit | No Mortality | No Mortality |
| N. leedsi | Binomial<br>Moving Average<br>Probit | No Mortality | No Mortality |

[a] Since more than half the organisms died in this lowest test concentration (10%), the $LC_{50}$ is < 10%.

[b] Test invalidated due to excessive control mortality (> 10%).

[c] 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 9. Results of Chemical Analyses Performed on Recovered Water collected from the Jacksonville NAS Sewage Treatment Plant, NPDES #FL0000957, on 12 January, 1987.

## Organics

| | |
|---|---|
| Acid Extractables: | N.D. |
| Base Neutral Extractables: | N.D. |
| Pesticide Extractables: | Endosulfan (< 0.03 ug/l) |
| | Several unidentified compounds |

## Metals

| | |
|---|---|
| Antimony | N.D. |
| Arsenic | N.D. |
| Beryllium | < 10 ug/l |
| Cadmium | 3 ug/l |
| Chromium | 56 ug/l |
| Copper | < 25 ug/l |
| Lead | -a |
| Mercury | N.D. |
| Nickel | N.D. |
| Selenium | N.D. |
| Silver | 1 ug/l |
| Thallium | N.D. |
| Zinc | 20 ug/l |

N.D. = None Detected
[a] Lead analysis invalidated due to blank contamination.

## RESULTS AND DISCUSSION

The sample of recovered water colleted from the Jacksonville NAS facility, consisting of both domestic and industrial components, proved to be acutely toxic to Ceriodaphnia dubia, Daphnia pulex, and Notropis leedsi, producing 24 hour $LC_{50}$'s of < 10%, and 17.9%, respectively (Table 8). This rapid and extreme toxicity was mostly due to the residual chlorine (1.0 mg/l) measured in the sample. This same sample, after de-chlorination, was not toxic to the test organisms. These results were very similar to previous results of DER bioassays of this facility, dating back to 1981, in which chlorine was mostly responsible for any observed toxicity.

The ammonia measured in the samples (up to 0.08 mg/l unionized) was insufficient to have contributed to the toxicity. However, chemical analyses (Table 9), indicated the presence of the pesticide endosulfan, several unidentified compounds, and the metals cadmium, chromium, silver, and zinc in the sample. These chemical components probably contributed to the toxicity noted in the Jacksonville NAS sample.

## 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.

The two basic concepts utilized in the standards are acute and chronic toxicity (defined in 17-3.021(1) and (4), respectively). Simplifying the actual F.A.C. language somewhat, acute toxicity is defined as the presence of a substance in a concentration one-third as great as that concentration which kills one-half of the test organisms in 96 hours, i.e., one-third of the 96 hour $LC_{50}$. Chronic toxicity is similarly defined as onetwentieth of the 96 hour $LC_{50}$. Therefore, if a test solution kills one- half or more of a group of test organisms in 96 hours, it must contain at least three times the defined "acutely toxic" concentration and at least twenty times the "chronically toxic" concentration of some substance or substances. If a test solution causes that same mortality in less than 96 hours, (for example, in 24 or 48 hours), it is at least as toxic as the example given above, and actually is probably considerably more toxic.

The particular restrictions applied to a given discharger depend upon the existence of a mixing zone, whetheras a specific part of the permit or based on the existence of a currently valid permit predating the language in Chapter 17-4 requiring mixing zone designation. In the absence of a mixing zone, the general criteria for surface waters [17-3.061] are applicable at all points, beginning at the point of discharge. This section prohibits the discharge of substances in concentrations which are chronically toxic [17-3.061(2)(p)]; that is, the concentrations cannot exceed one-twentieth of the 96 hour $LC_{50}$ as discussed above. If a mixing zone does exist, then this chronic toxicity standard is only applicable from the edge of the mixing zone and beyond. Within the mixing zone, the less stringent acute toxicity standard of the minimum criteria for all waters [17-3.051(1)(d)] applies: one-third of the 96 hour $LC_{50}$. The acute toxicity standard does not apply, however, to each point within a mixing zone not designated for dredge and fill permitting, but rather to the average of a number of points within such a mixing zone, based on the provisions of the surface water mixing zones section of Chapter 17-4 [17-4.244(4)]. At the same time, though, this same rule prohibits waste concentrations within such a mixing zone (i.e., at any point within the mixing zone), from exceeding the 96 hour $LC_{50}$. Therefore, at any point within such a mixing zone, including the point of discharge, one-half of the test organisms cannot be killed in 96 hours. Within a mixing zone designated for dredge and fill permitting, the acute toxicity standard (one-third of the 96 hour $LC_{50}$) applies at all points, including the point of discharge.

From the above discussion, it should be clear that if the least stringent toxicity standard, the 96 hour $LC_{50}$, is exceeded at any point, from the point of discharge outwards, by any discharger, then a violation of the standards has occurred. This is an important consideration since the test procedure here reported involves the measurement of the 50% lethal concentration, and not some fraction of that concentration. Any discharger required to meet the one-twentieth of the 96 hour $LC_{50}$ standard is therefore being tested very leniently. Any reduction in the test period from 96 hours provides an added degree of leniency to a discharger.

A number of different dilutions were examined in order to determine by how much, if any, the discharge exceeds the 96 hour $LC_{50}$. For example, if the sample, after two-fold dilution with diluent (a 33% concentration), kills one-half of the test organisms after 96 hours, then the sample is three times as toxic as is allowed by that standard. Similarly, if a 5% solution of the sample kills 50% of the organisms, the sample is twenty times as toxic as is allowed. Toxicities are reported here in that manner, i.e., in terms of the percentage dilution of the test sample. The smaller the percentage which is reported means a correspondingly greater toxicity.

Note that even if a discharger receives a very large mixing zone not designated for dredge and fill permitting, the regulations require that the maximum toxicity at any point within the mixing zone be restricted to the 96 hour $LC_{50}$. The results here reported are based on actual samples, representative of the discharge concentration which enters waters of the state at the point of discharge. If it is demonstrated that, under the conditions existing at the point of discharge, concentrations will be less than at the actual sampling point (if the two points are different), the reported bioassay results will reflect the reduced discharge concentration.

With respect to the species of organisms used in these tests, the language of the F.A.C. indicates that they should be "significant to the indigenous aquatic community". All species used in these tests are significant to the indigenous aquatic community since they are sensitive to toxic substances which may be expected to impact the indigenous community.

The toxicity reported here is in excess of allowable limits.

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