BIOASSAYS OF FLORIDA TILE - SIKES CORPORATION

LAKELAND, POLK COUNTY, FLORIDA

NO NPDES PERMIT

Biological Section
Bureau of Water Analysis
July 19, 1982

BIOASSAYS OF FLORIDA TILE - SIKES CORPORATION

LAKELAND, POLK COUNTY, FLORIDA

NO NPDES PERMIT

Biological Section
Bureau of Water Analysis
July 19, 1982

EXECUTIVE SUMMARY

Florida Tile - Sikes Corporation, Lakeland, on 25 to 27 May, 1982

The sample of potential hazardous waste effluent from this facility, a manufacturer of ceramic tiles, was acutely toxic to the test organisms, Daphnia pulex and Heterandria formosa. The chlorinated effluent produced 48 hour LC_{50} values of <5.6% and 70%, respectively, for Daphnia and Heterandria. A de-chlorinated subsample of the effluent produced 48 hour LC_{50} values of 35% to 38% for Daphnia. The cause of the toxicity in the chlorinated effluent sample may be attributed to the presence of total residual chlorine. Chemical analyses indicated the presence of significant concentrations of copper (31 µg/l), lead (40 µg/l) and zinc (88 µg/l) which may have acted synergistically and contributed to the observed toxicity.

Introduction

The Florida Tile - Sikes Corporation is located at 608 Prospect St. in Lakeland, Polk County, Florida (Fig. 1). This facility manufactures glazed ceramic wall and floor tiles. Wastewater from this operation is contained onsite in unlined waste ponds and there is supposedly no discharge of waste material. Recently, however, Mr. John Kolek of DER's Tampa Office was alerted to the fact that waterfowl in Lake Wire, adjacent to the Florida Tile facility, were being adversely affected by some unknown factor. Analyses of sediments from Lake Wire determined the presence of extremely high concentrations of lead, thereby classifying the sample as a hazardous waste. Suspected sources of the lead contamination were leachate from Florida Tile's unlined waste ponds and from a storm drain emptying to Lake Wire that was believed to originate from the Florida Tile facility.

In March, 1982, Mr. Kolek requested a toxicity bioassay be performed on liquid entering Lake Wire from the storm drain. This test produced a 48 hour LC_{50} of approximately 30% for the test organism, Daphnia pulex. The sample, however, arrived late in Tallahassee, thereby invalidating the test. Also, since the sample was collected from the end of the storm drain in Lake Wire, other factors could have been present. Therefore, the Biological Section recommended that the bioassay be repeated, and that the sample be taken from the storm drain as close as possible to the Florida Tile facility to preclude interference by other factors. DER personnel were not permitted on Florida Tile property for either inspection of their waste treatment facility or for sample collection.

In May, 1982, Mr. Matt McCann of the Tampa Office collected a sample of liquid from a manhole access point in the storm drain nearest the Florida Tile facility. He also noted that the storm drain was coming from the facility and that the flow was approximately 10-15 gpm. To further evaluate the existence and extent of toxicity of this effluent to the biota of the receiving waters, the Biological Section performed three static acute toxicity bioassays on this grab sample, on 25 to 27 May, 1982. The results of these tests are presented in this report.

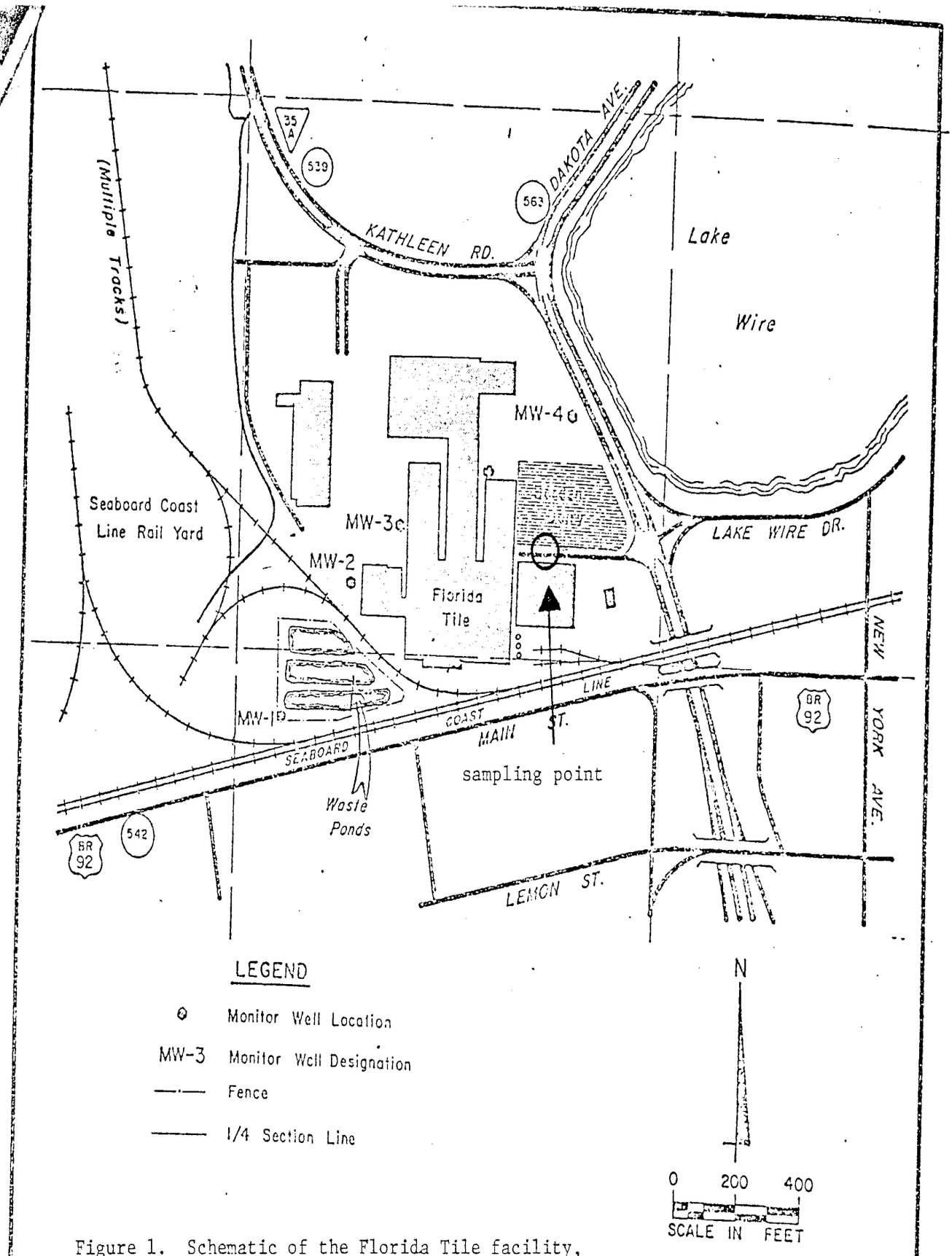


Figure 1. Schematic of the Florida Tile facility, Lakeland, Polk County, Florida, including location of storm drain sampling point.

RCF

METHODS AND MATERIALS

Test methods used to conduct these static bioassays are extensively described by Peltier (1978). The organisms selected for this study include a species recommended by Peltier (1978) and the U.S. EPA (1978). Additionally, the Biological 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, and are kept at a constant temperature of 22° plus or minus 1°C. The aquaria are cleaned weekly by siphoning off half the water along with the detritus and excess organisms. They are then refilled 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.

Heterandria formosa (least killifish) - Approximately 100 adult least killifish were obtained from a wild population in the St. Marks Wildlife Refuge (Wakulla County, Florida) during August, 1980. The fish are maintained in two and one-half to 15 gallon all glass aquaria equipped with a flow through water exchange system. Water for this system is unchlorinated and is pumped from a deep well near the DER laboratory. The least killfish are reared at 22° plus or minus 2°C under a 16:8 hour light-dark cycle and are fed freshly hatched Artemia (brine shrimp) twice daily. New born fish are initially maintained in the adult tanks, but are removed at weekly intervals, thereby segregating fish into even age groups. The fish used in this test were 1 to 2 weeks old.

To ensure the uniform sensitivity of these test organisms to toxic substances, the Biological Section conducts monthly standard reference toxicant bioassays using sodium lauryl sulfate as the reference toxicant.

Test Methods

Three 48 hour static acute toxicity bioassays were performed on a grab sample of effluent from a storm drain nearest the Florida Tile facility. The tests were performed from 1110 hours on 25 May to 1120 hours on 27 May, 1982.

The sample was collected by Mr. Matt McCann of DER's Southwest District at 1130 hours on 24 May, 1982, using standard wastewater sampling procedures. Separate samples were obtained for chemical and biological testing. Samples for metal analysis were preserved with HNO_3. Samples for organic chemical analysis and for the bioassays were unpreserved. The samples were iced and transported to the DER laboratory in Tallahassee for analysis within 24 hours of collection. Upon arrival in Tallahassee, the bioassay personnel determined the presence of a residual chlorine concentration of approximately 0.7 ppm in the effluent.

To differentiate between the toxicity resulting from chlorine and the toxicity associated with some other component of the effluent, the Biological Section adopted the following test procedure:

Test #1. Chlorinated Effluent (Tables 2 and 3). This test was performed on the actual effluent sample using Daphnia pulex and Heterandria formosa.

Test #2. De-chlorinated Effluent (Table 4). This test, using Daphnia pulex, was performed on a subsample of the chlorinated effluent sample in which the chlorine had been neutralized. De-chlorination was accomplished by adding a calculated amount of 0.025 N sodium thiosulfate. This test was intended to evaluate the toxicity of the effluent without the added toxic effects of chlorine.

Freshwater for dilution of the effluent sample 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 that recommended by APHA (1980).

Two replicates, each with six concentrations and a control, were used to test with the water flea, Daphnia pulex. Flint glass jars of 250 ml capacity were used as test chambers, and 200 ml of the appropriate test concentration was placed in each chamber. 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 five concentrations and a control, were used for the Heterandria formosa (least killifish) 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.

Dissolved oxygen, pH, temperature, and the number of live organisms were recorded for each test chamber at 0 hours, 24 hours, and 48 hours. Total alkalinity and total hardness were recorded at the beginning of the test for the controls and the 100% effluent concentration. The specific conductance of each test concentration was recorded at the beginning and end of the test. Ammonia concentrations, including total and unionized fractions (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 determined 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

Daphnia pulex (water flea)

Concentration	Dilution Water	Effluent	Total Volume
Control	200 ml	---	200 ml
5.6%	188.8 ml	11.2 ml	200 ml
10.0%	180 ml	20 ml	200 ml
18.0%	164 ml	36 ml	200 ml
32.0%	136 ml	64 ml	200 ml
56.0%	88 ml	112 ml	200 ml
100.0%	--	200 ml	200 ml

Heterandria formosa (least killifish)

Concentration	Dilution Water	Effluent	Total Volume
Control	500 ml	---	500 ml
10.0%	450 ml	50 ml	500 ml
18.0%	410 ml	90 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, *Daphnia pulex*, static acute toxicity bioassay of Florida Tile, Lakeland, Polk County, Florida, on 25 to 27 May, 1982.

Test #1 Chlorinated Sample[a]

Conc. or %	Number of Live Organisms			Dissolved Oxygen (mg/ℓ)			pH			Total Alkalinity (mg/ℓ as $CaCO_3$)		Total Hardness (mg/ℓ as $CaCO_3$)		Temperature (°C)			NH_3/NH_3+NH_4 (mg/ℓ)		Conductivity (µmhos)	
	0	24	48	0	24	48	0	24	48	0	48	0	48	0	24	48	0	48	0	48
Control	10	10	10	9.0	9.5	9.2	7.8	8.1	8.3	119		118		21.9	18.5	19.8	0/0	---		
Control	10	10	10	9.0	9.7	9.2	7.8	8.2	8.3										192	200
5.6%	10	9	0	9.0	9.7	9.2	7.8	8.2	8.3					21.8	18.5	19.8				
5.6%	10	3	1	9.0	9.8	9.2	7.8	8.2	8.3										200	202
10.0%	10	0	0	8.9	9.8	--b	7.8	8.2	--[b]					21.7	18.5	--[b]				
10.0%	10	0	0	8.9	9.8	--	7.8	8.2	--										202	--[b]
18.0%	10	0	0	8.9	9.9	--	7.9	8.2	--					21.7	18.5	--				
18.0%	10	0	0	8.9	9.9	--	7.9	8.2	--										210	---
32.0%	10	0	0	8.8	9.9	--	7.9	8.2	--					21.6	18.5	--				
32.0%	10	0	0	8.8	9.9	--	7.9	8.2	--										212	---
56.0%	10	0	0	8.6	9.9	--	7.9	8.2	--					20.9	18.7	--				
56.0%	10	0	0	8.6	9.9	--	7.9	8.2	--										225	---
100.0%	10	0	0	8.0	9.9	--	7.9	8.2	--					21.0	19.0	--	0/0	---		
100.0%	10	0	0	8.0	9.9	--	7.9	8.2	--	135		133							245	---

Remarks:

[a] Total Residual Chlorine: 0.65 mg/l (HACH DPD Method)

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

Table 3. Data recorded during 48 hour, *Heterandria formosa*[a], static acute toxicity bioassay of Florida Tile, Lakeland, Polk County, Florida, on 25 to 27 May, 1982.

Test #1 Chlorinated Sample[b]

Conc. or %	Number of Live Organisms			Dissolved Oxygen (mg/ℓ)			pH			Total Alkalinity (mg/ℓ as $CaCO_3$)		Total Hardness (mg/ℓ as $CaCO_3$)		Temperature (°C)			NH_3/NH_3+NH_4 (mg/ℓ)		Conductivity (µmhos)	
	0	24	48	0	24	48	0	24	48	0	48	0	48	0	24	48	0	48	0	48
Control	5	5	5	8.9	9.0	8.8	7.8	8.0	8.1	119		118		21.2	18.8	19.5	0/0	---		
Control	5	5	5	8.9	9.0	8.8	7.8	8.0	8.1										210	210
10.0%	5	5	5	8.9	9.1	8.9	7.8	8.1	8.2					21.1	18.6	19.4				
10.0%	5	5	5	8.9	9.1	8.9	7.8	8.1	8.2										215	215
18.0%	5	5	5	8.8	9.1	9.0	7.9	8.1	8.2					21.1	18.6	19.4				
18.0%	5	5	5	8.8	9.1	9.0	7.9	8.1	8.2										220	220
32.0%	5	5	5	8.8	9.1	9.1	7.9	8.1	8.2					21.1	18.5	19.4				
32.0%	5	5	5	8.8	9.2	9.1	7.9	8.1	8.2										225	225
56.0%	5	5	4	8.5	9.2	9.2	7.9	8.1	8.2					21.0	18.5	19.4				
56.0%	5	5	5	8.5	9.2	9.2	7.9	8.1	8.2										235	235
100.0%	5	0	0	8.3	9.3	8.4	7.9	8.1	8.1	135		133		21.2	18.7	19.5	0/0	---		
100.0%	5	0	0	8.3	9.3	8.4	7.9	8.1	8.1										260	260

Remarks:

[a] Weight of fish: 0.0112 g each, average of 10 fish.
[b] Total Residual Chlorine: 0.65 mg/l (HACH DPD Method).

Table 4 . Data recorded during 48 hour, Daphnia pulex, static acute toxicity bioassay of Florida Tile, Lakeland, Polk County, Florida, on 25 to 27 May, 1982.

Test #2 De-Chlorinated Sample[a]

Conc. or %	Number of Live Organisms			Dissolved Oxygen (mg/ℓ)			pH			Total Alkalinity (mg/ℓ as $CaCO_3$)		Total Hardness (mg/ℓ as $CaCO_3$)		Temperature (°C)			NH_3/NH_3+NH_4 (mg/ℓ)		Conductivity (µmhos)	
	0	24	48	0	24	48	0	24	48	0	48	0	48	0	24	48	0	48	0	48
Control	10	10	9[b]	8.7	9.5	9.2	7.8	8.2	8.3	119		118		21.1	18.5	19.7	0/0	---		
Control	10	10	9[b]	8.7	9.5	9.2	7.8	8.2	8.3										200	208
5.6%	10	10	10	8.8	9.5	9.2	7.8	8.2	8.3					21.1	18.3	19.6				
5.6%	10	10	10	8.8	9.5	9.2	7.8	8.2	8.3										210	210
10.0%	10	10	10	8.8	9.5	9.3	7.8	8.2	8.3					21.2	18.3	19.5				
10.0%	10	9	9	8.8	9.5	9.3	7.8	8.2	8.3										212	211
18.0%	10	9	8	8.8	9.6	9.3	7.9	8.2	8.3					21.3	18.5	19.5				
18.0%	10	9	9	8.8	9.5	9.3	7.9	8.2	8.3										215	218
32.0%	10	9	8	8.8	9.6	9.3	7.9	8.2	8.3					21.3	18.7	19.5				
32.0%	10	8	5	8.8	9.6	9.3	7.9	8.2	8.3										222	229
56.0%	10	6	1	8.7	9.6	9.3	8.0	8.2	8.3					21.6	18.5	19.6				
56.0%	10	7	3	8.7	9.7	9.3	8.0	8.2	8.3										235	238
100.0%	10	7	1	8.4	9.7	9.1	8.0	8.2	8.3	135		133		22.0	19.2	19.6	0/0	---		
100.0%	10	8	0	8.4	9.7	9.1	8.0	8.2	8.3										260	260

Remarks:

[a] Total Residual Chlorine: <0.02 mg/l (HACH DPD Method)

[b] One organism not found.

Table 5. LC_{50} Data for Bioassays of Florida Tile, Lakeland, on 25 to 27 May, 1982.

Organism	Method of Data Analysis	24 LC_{50} and 95% Confidence Intervals	48 Hour LC_{50} and 95% Confidence Intervals
Daphnia pulex	Binomial	6.0% (0 and 10)	<5.6%, b
Chlorinated	Moving Average	a	<5.6%, b
Sample	Probit	a	<5.6%, b
H. formosa	Binomial	74.8% (56 and 100)	70.4% (56 and 100)
Chlorinated	Moving Average	a	a
Sample	Probit	a	a
Daphnia pulex	Binomial	a	38.3% (18 and 56)
De-chlorinated	Moving Average	a	35.7% (28 and 46)
Sample	Probit	200.7% (93 and 2967)	35.5% (29 and 44)

[a] Due to the pattern of mortality, an LC_{50} value cannot be calculated using this method of statistical analysis.

[b] Since almost all organisms (19 of 20) were dead in the lowest test concentration (5.6%), the LC_{50}'s <5.6%.

RESULTS AND DISCUSSION

The sample of effluent collected from the storm drain in front of the Florida Tile facility was extremely toxic to Daphnia pulex, with 24 and 48 hour LC_{50} values of 6.0% and <5.6%, respectively (Table 5). The residual chlorine measured in this sample (0.65 mg/l) was sufficient to have caused some, but not all, of the observed toxicity. This same effluent was also acutely toxic to Heterandria formosa, with 24 and 48 hour LC_{50} values of 74.8% and 70.4%, respectively (Table 5). The residual chlorine may have constributed to this toxicity. The de-chlorinated subsample of effluent was acutely toxic to Daphnia pulex, with 48 hour LC_{50} values of from 35.5% to 38.3% (Table 5). The results of these tests suggest that toxic factors other than chlorine may be present in this effluent. The 96 hour LC_{50}'s for these tests would be less than or equal to the 48 hour values.

The DER Chemistry Section has analyzed samples of this effluent for certain organic compounds and metals (Table 6). Of the organic compounds tested for, none were detected in the effluent. The analyses for metals, however, detected the presence of significant concentrations of copper (31µg/l), lead (40µg/l) and zinc (88µg/l). The concentrations of these metals, although individually sub-lethal (U.S. EPA 1976), may have acted synergistically and may have caused at least part of the observed toxicity.

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 (3), 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 one-twentieth 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 sustance 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, whether as 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)(o)); 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, but rather to the average of a number of points within 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 the mixing zone (i.e., at any point within the mixing zone), from exceeding the 96 hour LC_{50}. Therefore, at any point within a mixing zone, including the point of discharge, one-half of the test organisms cannot be killed in 96 hours.

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} standard. 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, 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 effluent samples, representative of the effluent 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, effluent 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.

Table 6. Results of Chemical Analyses Performed on Effluent from Florida Tile, Lakeland, collected 24 May, 1982

Organics

Parameter	Compound
Acid Extractables	N.D.
Base Neutral Extractables	N.D.
Pesticide Extractables	N.D.

Metals

Antimony	N.D.
Arsenic	<20 µg/l[a]
Beryllium	N.D.
Cadmium	N.D.
Chromium	N.D.
Copper	31 µg/l
Lead	40 µg/l
Mercury	N.D.
Nickel	N.D.
Selenium	N.D.
Silver	N.D.
Thallium	N.D.
Vanadium	<10 µg/l[a]
Zinc	88 µg/l

N.D. = None Detected

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

LITERATURE CITED

American Public Health Association. 1980. Standard Methods for the Examination of Water and Wastewater. 15th ed. 1134 p.

Peltier, W. 1978. Methods for measuring the acute toxicity of effluents to aquatic organisms. Environmental Research Laboratory. Athens, Georgia. EPA-600/4-78-012 (Revised July 1978). 51 p.

U.S. Environmental Protection Agency. 1976. Quality Criteria for Water. U.S. EPA, Washington, D.C. 256 p.

U.S. Environmental Protection Agency. 1978. Bioassay procedures for the Ocean Disposal Permit Program. Environmental Research Laboratory. Gulf Breeze, Florida. EPA-600/9-78-010. 121 p.