BIOASSAYS OF ALTON PACKAGING COMPANY

JACKSONVILLE, DUVAL COUNTY, FLORIDA

NPDES #FL0000892

SAMPLED 1/7/85

FILE COPY
DO NOT REMOVE

February 19, 1985

Biology Section
Division of Environmental Programs

BIOASSAYS OF ALTON PACKAGING COMPANY

JACKSONVILLE, DUVAL COUNTY, FLORIDA

NPDES #FL0000892

Biology Section
Bureau of Laboratories
February 19, 1985

EXECUTIVE SUMMARY

Alton Packaging Company, Jacksonville, NPDES #FL0000892, on 8 to 10 January, 1985.

The wastewater effluent from Alton Packaging Company was not toxic to the test organisms Daphnia pulex, Heterandria formosa, and Mysidopsis bahia. In the D. pulex bioassay, no significant mortality occurred. No mortality occurred in the H. formosa test. In the M. bahia test, 30% of the organisms died in the 100% concentration. This mortality was, however, insufficient to calculate an LC_{50}. Due to this wastewater's extremely low initial dissolved oxygen content, and to its high biochemical oxygen demand, the test chambers were aerated moderately throughout the test. This procedure significantly increased the pH and may have rendered the effluent less toxic. The total ammonia concentration (.15 mg/l) measured in the wastewater sample was insignificant. Chemical analyses were not performed since the sample was not toxic.

INTRODUCTION

The Alton Packaging Company is located at 1915 Wigmore Street, Jacksonville, Duval County, Florida (Figure 1). This paper industry facility uses pine wood to produce approximately 707 tons per day of Kraft pulp and linerboard. In the production process, 5 MGD of wastewater is generated which is discharged to the St. Johns River. Prior to discharge, the wastewater is treated by retention in a settling pond and by a Unox® treatment system. The wastewater is also fertilized with ammonia to promote the growth of microorganisms to aid in the degradation of wood fibers present in the wastewater. Sludges are separated from the wastewater prior to discharge.

This facility has, on occasion, been in violation for exceeding certain NPDES permit limitations. Additionally, the FDER Biology Section had previously performed a toxicity test on effluent from this facility, in September, 1981. The results of that test indicated that the effluent was extremely toxic to the test organism, Mysidopsis bahia, with a 48 hour LC_{50} of 15.7% (FDER 1981). Toxicity was attributed, in part, to the large concentrations of total and unionized ammonia determined in the sample at that time. However, Alton Company officials claimed that an ammonia spill occurred the same day that the sample was collected.

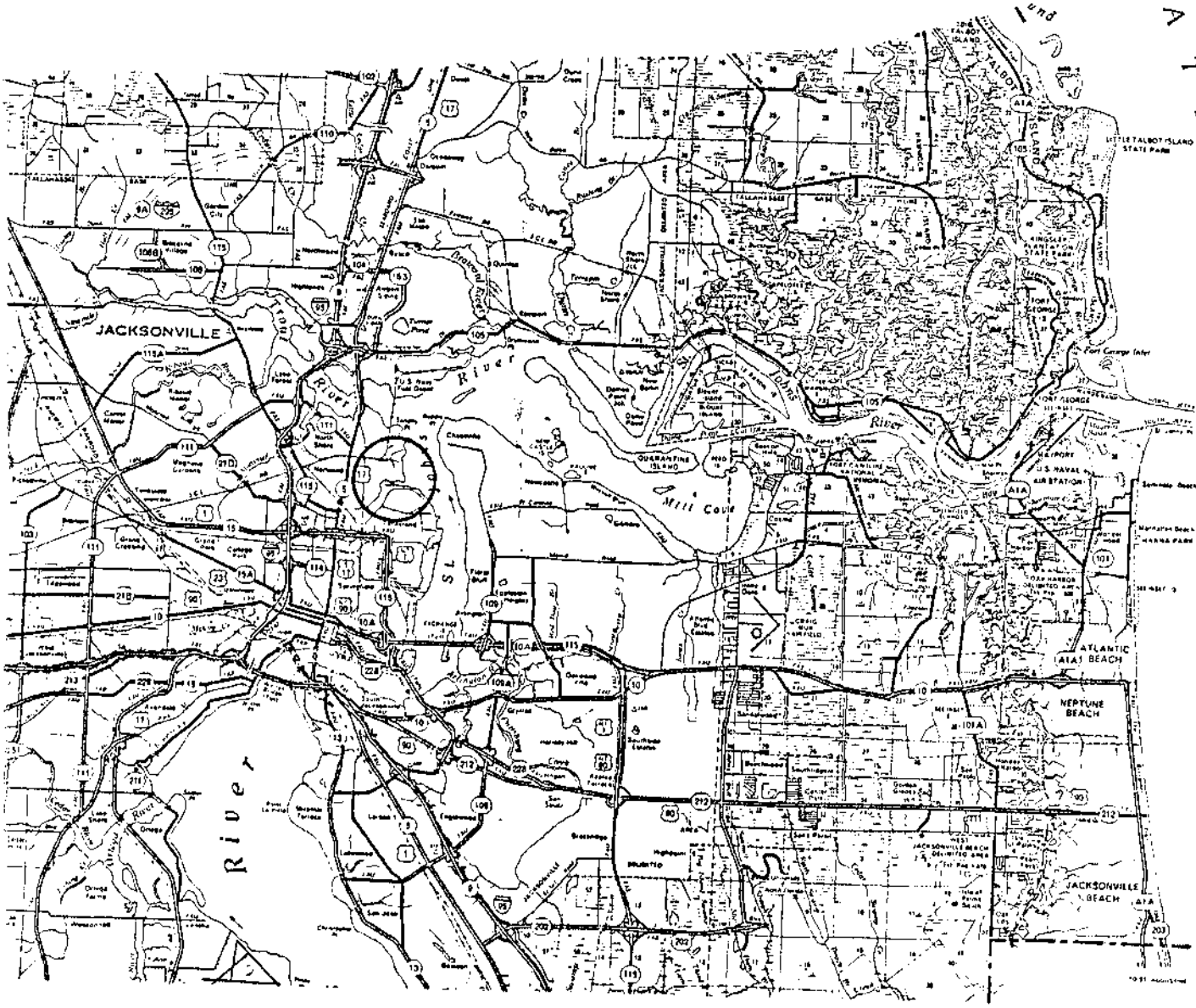
Effluent from this facility was retested, in March, 1982, again using Mysidopsis bahia as the test organism. Results of that test, however, were invalidated due to a large biochemical oxygen demand exerted by the effluent that depleted the dissolved oxygen in the test chambers and resulted in organism mortality. Therefore, that test was inconclusive in determing toxic aspects of the effluent. Ammonia was not detected in the effluent at that time (FDER 1982a).

Since the March, 1982, test was invalidated due to depleted dissolved oxygen, two additional bioassays were performed in July, 1982, using Mysidopsis bahia and Daphnia pulex as test organisms. The higher test concentrations in these bioassays were continuously aerated to negate the effects of the effluent's high BOD. The M. bahia test produced an LC_{50} of 141.7%, a violation of the Chapter 17-3 acute toxicity standard. The D. pulex test was invalidated due to missing organisms in the extremely opaque effluent (FDER 1982b). However, after negotiating with DER's Jacksonville office, Alton Company was granted a mixing zone, thereby bringing the effluent into compliance with regard to acute toxicity as stated in the Chapter 17-4 mixing zone criteria.

Due to the history of toxicity at this facility and the recent granting of a mixing zone to bring this toxicity into compliance, additional bioassay testing was requested by the Northeast District. Two 48 hour static bioassays were performed on a sample of Alton's effluent on 24 to 26 May, 1983, using Mysidopsis bahia and Heterandria formosa as test organisms. The results of these tests indicated 30% mortality of M. bahia in the 100% concentration, while no H. formosa mortality was noted (FDER 1983a). All test chambers were continuously aerated.

To continue to evaluate the toxicity of this effluent, three 48 hour static acute toxicity bioassays were performed on 27 to 29 March, 1984, using Mysidopsis bahia, Notropis leedsi and Daphnia pulex as test organisms. The M. bahia produced a valid 24 hour LC_{50} of 87.7%. However, at 48 hours, the dissolved oxygen in several concentrations had decreased to below recommended levels and additional organism mortality was not used in further LC_{50} calculations. In the D. pulex bioassay, 25% of the organisms died in the 100% concentration which was insufficient to calculate an LC_{50}. In the N. leedsi bioassay, only 10% of the organisms had died in the 100% concentration at 24 hours. At 48 hours, however, similar to the M. bahia test, insufficient dissolved oxygen was noted in the test chambers, and additional organism mortality was not used to calculate an LC_{50} (FDER 1984).

To again evaluate the toxicity of this discharge, three 48 hour static acute toxicity bioassays were performed on 8 to 10 January, 1985. Organisms selected for these tests were again Mysidopsis bahia, Heterandria formosa and Daphnia pulex. The results of these latest tests are presented in this report. Those involved in performing the tests and in preparation of this report were: R. B. Frydenborg, M. L. Moultrie, and M. L. Roll.



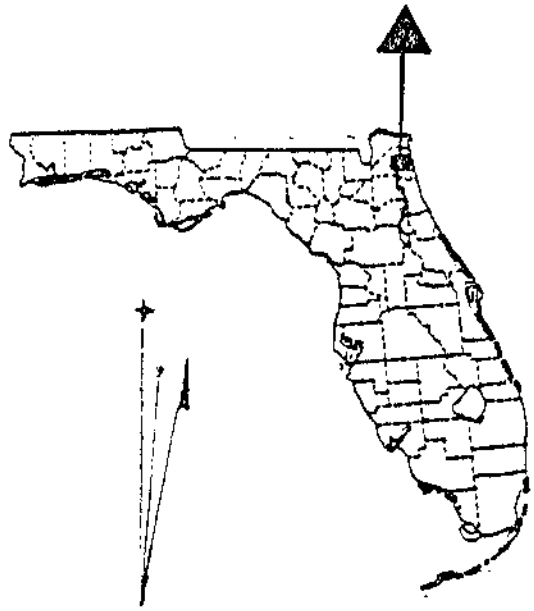


Figure 1. Portions of Jacksonville, Duval County, Florida, including approximate location of Alton Packaging Company.

METHODS AND MATERIALS

Test methods used to conduct these static bioassays are extensively described by Peltier (1978) and the FDER Biology Section (1983). The organisms selected for this study include species recommended by Peltier (1978), the U.S. EPA (1978), and the FDER Biology Section (1983). 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° plus or minus 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.

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 killifish 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) and Tetra-Min® twice daily and once daily on weekends. New born fish are initially maintained in the adult tanks, but are transferred to separate aquaria at weekly intervals, thereby segregating fish into even age groups. The fish used in this test were 3-4 weeks old.

Mysidopsis bahia (mysid shrimp) - In September, 1978, approximately 300 adult mysid shrimp were obtained from the EPA laboratory in Gulf Breeze, Florida. The mysids are cultured under a 16:8 hour light-dark cycle in 10 and 15 gallon all-glass aquaria equipped with undergravel filtration and aeration. The mysid shrimp are cultured over a wide range of salinities (10-30 ppt) to minimize acclimation periods to low or high salinity effluents. Seawater, used for culturing the test organisms and as diluent in the bioassays, is prepared by mixing artificial sea salts (Forty Fathoms®) and well water to obtain a final salinity of 20-30 ppt. Freshwater, for dilution of the seawater as necessary, is obtained from a deep well near the DER laboratory. The mysids are fed twice daily and once daily on weekends with freshly hatched Artemia (brine shrimp) and are kept at a constant temperature of 23° plus or minus 3°C. Adult or large juvenile mysids were used in this 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

Three 48 hour static acute toxicity bioassays were performed on a sample of Alton's effluent. The tests were performed from 1110 hours on 8 January to 1200 hours on 10 January, 1985.

The sample was collected by Mr. Martin Zenas of DER's Northeast District at 1400 hours on 7 January, 1985, 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.

Freshwater for dilution of the effluent sample for the D. pulex and H. formosa tests 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, each with three concentrations and a control, were used to test 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 water fleas 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 this test.

Two replicates, of three 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.

Saltwater for dilution of the effluent sample for the Mysidopsis bahia test was prepared from artificial sea salts (salinity = 31.5 ppt). For test concentrations of 56% and below, this diluent was mixed directly with the effluent, achieving a range of final salinities (16.2 ppt to 29.0 ppt). For the 100% effluent test concentrations, freshly autoclaved artificial sea salts were added until salinities of approximately 7 ppt were achieved (the lower limit of mysid shrimp salinity tolerance). Four control groups (Tables 4 and 5) were used to ensure that any mortality in the test organisms was not the result of toxic properties associated with the diluent, the artificial sea salts, or the salinity range. The control groups included:

1) Diluent Control - Artificial seawater (31.5 ppt). This represents the highest concentration of artificial seawater used in the bioassay, and tests for toxicity in the diluent. This is also the highest salinity used in the bioassay, and therefore tests the tolerance of the organisms to this salinity.

2) Salinity Control - Artificial seawater, adjusted with well water to approximately 7 ppt salinity. This tests for tolerance of the organisms to the lowest salinity used in the bioassay.

3) pH Adjusted Effluent Control - Effluent (100%) which has been adjusted to a salinity of approximately 7 ppt using artificial sea salts, and which has then been pH adjusted using hydrochloric acid to the original pH of the effluent at the time of sampling. This test ensures that toxicity, or lack of toxicity, observed in the bioassay is representative of the wastewater during discharge and is not merely due to a change in pH caused by the sea salts.

4) Acid Control - Well water, adjusted with artificial sea salts to a salinity of approximately 7 ppt, with an amount of hydrochloric acid added needed to equal the pH achieved in the pH Adjusted Effluent Control. This tests for toxicity in the pH Adjusted Effluent Control due to the hydrochloric acid.

The concentration series selected for this study (Table 1) is that recommended by APHA (1980). Two replicates of each test concentration and of the controls were used to test with Mysidopsis bahia. Immediately prior to the bioassays, a suitable number of mysids were transferred by net from the 10 ppt, 15 ppt, 20 ppt, 25 ppt and 30 ppt salinity aquaria to culture dishes for observation and selection of individuals for the tests. Individual mysid shrimp were siphoned using a glass tube, carefully examined for injury, and transferred to wide mouth quart jars, each containing 500 ml of the appropriate concentration. For each concentration, the mysid shrimp were pre-acclimated to within 4 ppt salinity. Five organisms were loaded per test chamber, giving ten per concentration. Based on the recommendations of EPA (1978), the mysids were fed brine shrimp (Artemia) nauplii at the rate of approximately 10 to 20 nauplii per mysid per day to minimize cannibalism.

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 or salinity (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 through 5.

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

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
50.0%	100.0 ml	100.0 ml	200 ml
100.0%	--------	200.0 ml	200 ml

Heterandria formosa (least killifish)

Mysidopsis bahia (mysid shrimp)

Concentration	Dilution Water	Effluent	Total Volume
Control	500 ml	------	500 ml
10.0%[a]	450 ml	50 ml	500 ml
18.0%	410 ml	90 ml	500 ml
32.0%	340 ml	160 ml	500 ml
50.0%[a]	250 ml	250 ml	500 ml
56.0%	220 ml	280 ml	500 ml
100.0%[a]	------	500 ml	500 ml

[a] These concentrations were used in the *H. formosa* test.

Table 2. Data recorded during 48 hour, Daphnia pulex, static acute toxicity bioassays of Alton Packaging Company, FL0000892, Jacksonville, Duval County, Florida, on 8 to 10 January, 1985.

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	7.6 [c]	9.0	8.4	7.8	7.9	7.7	20.3	19.6	22.3		
Control	10	10	9	7.7 [c]	9.0	8.4	7.8	7.9	7.7				212	245
10.0%	10	10	10	7.6	8.1	8.0	7.5	7.9	7.8	20.4	19.8	22.4		
10.0%	10	10	10	7.6	8.1	8.0	7.5	7.9	7.8				440	430
50.0%	10	- [a]	7/8 [b]	6.7 [c]	8.6	8.1	7.2	8.2	8.1	20.4	19.2	23.1		
50.0%	10	-	4/6 [b]	6.7 [c]	8.6	7.8	7.2	8.2	8.1				1310	1250
100.0%	10	-	10	5.7 [c]	7.6	7.7	7.0	8.1	8.1	20.4	19.2	22.5		
100.0%	10	-	8	5.7 [c]	8.2	8.0	7.0	8.2	8.2				2300	2400

Control Water

NH_3(as NH_3)(mg/l) 0

Total Alkalinity (mg/l $CaCO_3$) 73

Total ammonia (as N)(mg/l) .07

Total Hardness (mg/l $CaCO_3$) 107

100% Effluent

NH_3(as NH_3)(mg/l) 0

Total Alkalinity (mg/l $CaCO_3$) 178

Total ammonia (as N)(mg/l) .15

Total Hardness (mg/l $CaCO_3$) 224

[a] Organisms could not be counted at 24 hours due to turbidity of sample.
[b] Seven out of eight and four out of six counted alive respectively, remaining organisms were not found.
[c] These chambers were aerated to the values shown prior to loading the organisms and were aerated moderately throughout the 48 hour test.

Table 3. Data recorded during 48 hour, Heterandria formosa, static acute toxicity bioassays of Alton Packaging Company, FL0000892, Jacksonville, Duval County, Florida, on 8 to 10 January, 1985.

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	7.9 [a]	8.7	8.6	7.8	8.1	8.1	20.4	19.2	22.1			Control Water
Control	5	5	5	7.9 [a]	8.7	8.6	7.8	8.1	8.1				212	331	NH_3(as NH_3)(mg/l) 0 — Total Alkalinity (mg/l $CaCO_3$) 73
10.0%	5	5	5	7.7	7.2	7.5	7.6	7.9	7.9	20.5	19.2	21.8			
10.0%	5	5	5	7.8	7.2	7.5	7.6	7.9	7.9				445	470	
50.0%	5	5	5	6.6 [a]	8.5	8.5	7.2	8.3	8.2	20.4	19.1	21.8			Total ammonia (as N)(mg/l) .07 — Total Hardness (mg/l $CaCO_3$) 107
50.0%	5	5	5	6.6 [a]	8.5	8.6	7.2	8.3	8.4				1350	1400	
100.0%	5	5	5	5.5 [a]	7.8	8.2	7.0	8.2	8.3	20.5	19.5	22.2			
100.0%	5	5	5	5.5 [a]	8.1	8.3	7.0	8.3	8.3				2400	2450	
															100% Effluent
															NH_3(as NH_3)(mg/l) 0 — Total Alkalinity (mg/l $CaCO_3$) 178
															Total ammonia (as N)(mg/l) .15 — Total Hardness (mg/l $CaCO_3$) 224

[a] These chambers were aerated to the values shown prior to loading the organisms and were aerated moderately throughout the 48 hour test.

Table 4. Data recorded during 48 hour, Mysidopsis bahia, static acute toxicity bioassay of Alton Packaging Company, FL0000892, Jacksonville, Duval County, Florida, on 8 to 10 January, 1985.

Concentration or %	Number of Live Organisms			Dissolved Oxygen (mg/l)			pH			Temperature (°C)			Salinity (ppt)		
	0	24	48	0	24	48	0	24	48	0	24	48	0		48
Diluent Control	5	5	5	7.3 [a]	7.0	7.2	8.3	8.4	8.6	19.5	19.1	21.6			
Diluent Control	5	5	5	7.3 [a]	7.0	7.3	8.3	8.5	8.5				31.5		32.5
Salinity Control	5	5	5	8.1	8.1	7.4	8.3	8.4	8.5	20.3	19.0	21.5			
Salinity Control	5	5	5	8.0	8.1	7.4	8.3	8.4	8.4				7.0		7.9
10.0%	5	5	5	7.2	6.4	6.2	8.3	8.3	8.2	19.7	19.2	21.8			
10.0%	5	4	4	7.2	6.4	6.2	8.3	8.3	8.4				29.0		30.5
18.0%	5	5	5	7.0	7.0	5.9	8.2	8.3	8.5	19.8	19.0	21.6			
18.0%	5	5	5	7.0 [a]	7.0	6.0	8.2	8.3	8.5				27.0		27.0
32.0%	5	5	5	6.9 [a]	7.5	7.5	8.1	8.4	8.6	19.8	19.0	21.0			
32.0%	5	5	5	6.9 [a]	7.5	7.5	8.1	8.5	8.6				23.5		24.8
56.0%	5	5	5	6.7 [a]	7.9	7.7	7.6	8.5	8.4	19.9	19.0	21.5			
56.0%	5	5	5	6.7 [a]	7.9	7.7	7.6	8.5	8.4				16.2		17.1
100.0%	5	5	3	5.6 [a]	8.1	8.0	7.2	8.6	8.4	20.0	19.2	21.8			
100.0%	5	4	4	5.6 [a]	8.1	8.0	7.2	8.5	8.4				7.0		8.0

Control Water

NH_3(as NH_3)(mg/l) 0

Total ammonia (as N)(mg/l) .07

Total Alkalinity (mg/l $CaCO_3$) 73

Total Hardness (mg/l $CaCO_3$) 107

100% Effluent

NH_3(as NH_3)(mg/l) 0

Total ammonia (as N)(mg/l) .15

Total Alkalinity (mg/l $CaCO_3$) 178

Total Hardness (mg/l $CaCO_3$) 224

[a] These chambers were aerated to the values shown prior to loading the organisms and were moderately aerated throughout the 48 hour test.

Table 5. Data recorded during 48 hour, Mysidopsis bahia, static acute toxicity bioassay of Alton Packaging Company, FL0000892, Jacksonville, Duval County, Florida, on 8 to 10 January, 1985.

Concentration or %	Number of Live Organisms			Dissolved Oxygen (mg/l)			pH			Temperature (°C)			Salinity (ppt)	
	0	24	48	0	24	48	0	24	48	0	24	48	0	48
Acid Control	5	5	3	8.1	8.1	7.4	6.9	7.8	8.2	20.1	19.1	21.7		
Acid Control	5	5	5	8.1	8.1	7.5	6.9	7.8	8.1				7.0	7.1
pH Adjusted Effluent	5	5	4	6.0 [a]	7.8	8.1	6.9	8.4	8.4	19.9	19.7	21.9		
pH Adjusted Effluent	5	5	5	6.0 [a]	8.0	8.1	6.9	8.4	8.4				7.8	8.1

Control Water

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

Total Alkalinity (mg/l $CaCO_3$): 73

Total ammonia (as N)(mg/l): .07

Total Hardness (mg/l $CaCO_3$): 107

100% Effluent

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

Total Alkalinity (mg/l $CaCO_3$): 178

Total ammonia (as N)(mg/l): .15

Total Hardness (mg/l $CaCO_3$): 224

[a] These chambers were aerated to the values shown prior to loading the organisms and were moderately aerated throughout the 48 hour test.

RESULTS AND DISCUSSION

These latest bioassay analyses on effluent from Alton Packaging Company indicated that the wastewater was not acutely toxic to the test organisms, Daphnia pulex, Heterandria formosa, and Mysidopsis bahia. In the D. pulex bioassay, no significant mortality occurred (Table 2), however due to the extremely opaque effluent (due to high concentrations of suspended solids) several organisms could not be found in some test chambers. No mortality occurred in the H. formosa test (Table 3), but only with difficulty could the fish be counted. In the M. bahia test, 30% of the organisms died in the 100% concentration (Table 4), however this mortality was insufficient to calculate an LC_{50}. These results were very similar to the May, 1983, test results, in which aerated Alton wastewater was also toxic to 30% of M. bahia.

Due to the history of low dissolved oxygen and a high biochemical oxygen demand in Alton's wastewater, the test chambers during these latest tests were aerated prior to loading the organisms and throughout the tests. Due to this aeration, a significant increase in pH was noted from 0 to 24 hours in most of the test concentrations, including the pH Adjusted 100% effluent (Tables 2-5). This negated the pH Adjustment and may have rendered the effluent less toxic to the test organisms. In a previous bioassay report (FDER 1984), it was suggested, and literature was cited, that paper mill wastewater is significantly less toxic at neutral or higher pH's. In the current tests, the pH of Alton's wastewater had increased from 6.9 (measured at sampling) to 8.2-8.6 by 24 hours due to the aeration. Therefore, it may be that Alton's wastewater was tested at a reduced toxic level during these tests. The total ammonia concentration (.15 mg/l) measured in the sample was insignificant. Since the wastewater was not acutely toxic, chemical analyses were not performed.

LITERATURE CITED

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

Florida Department of Environmental Regulation, Biology Section. 1981. Bioassay of Alton Box Board Company, Duval County, Florida. NPDES #FL0000892. FDER. 11 p.

_____. 1982a. Bioassay of Alton Box Board Packaging Company, Jacksonville, Duval County, Florida. NPDES #FL0000892. FDER. 11 p.

_____. 1982b. Bioassays of Alton Box Board Packaging Company, Jacksonville, Duval County, Florida. NPDES #FL0000892. FDER. 14 p.

_____. 1983a. Bioassays of Alton Box Packaging Company, Jacksonville, Duval County, Florida. NPDES #FL0000892. FDER. 10 p.

_____. 1983b. Quality Assurance Manual for Performing Acute Toxicity Tests. FDER. 22 p.

_____. 1984. Bioassays of Alton Packaging Company, Jacksonville, Duval County, Florida. NPDES #FL0000892. FDER. 18 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. 1978. Bioassay procedures for the Ocean Disposal Permit Program. Environmental Research Laboratory. Gulf Breeze, Florida. EPA-600/9-78-010. 121 p.