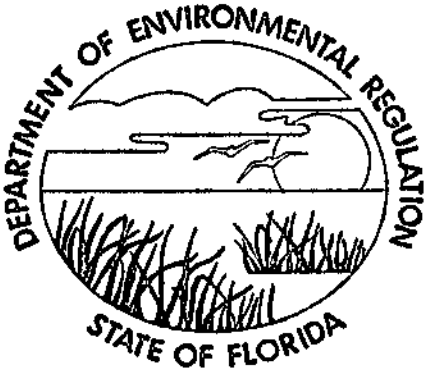


BIOASSAYS OF ALPHA CHEMICAL CORPORATION

POLK COUNTY, FLORIDA

NPDES #FL0029653

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OCTOBER 11, 1985

Biology Section
Division of Environmental Programs

BIOASSAYS OF ALPHA CHEMICAL CORPORATION

POLK COUNTY, FLORIDA

NPDES #FL0029653

Biology Section
Bureau of Laboratories and
Special Programs
October 11, 1985

EXECUTIVE SUMMARY

Alpha Chemical Corporation, NPDES #FL0029653, on 15 to 17 August, 1985.

The sample of effluent collected from the Alpha Chemical Corporation was not acutely toxic to the test organisms, Daphnia pulex, Ceriodaphnia dubia or Notropis leedsi. No mortality occurred in the D. pulex and N. leedsi 48 hour bioassays, and only random mortality was noted in the C. dubia 48 hour bioassay. Chemical analyses were not performed since the sample was not toxic.

INTRODUCTION

The Alpha Chemical Corporation is a polyester resin facility located in Kathleen, Polk County, Florida (Fig. 1). The facility manufactures approximately 70 tons per day of synthetic resins. For this production, approximately 0.4 MGD of groundwater is used, the majority of which is non-contact cooling water and boiler-feed water. The wastewater is discharged (after cooling in two ponds) through a drainage ditch to Swampy Bog, which eventually flows to the Hillsborough River. Boiler wash and caustic cleaning waters are evaporated in a concrete lined pond. The remaining process water and solid/semi-solid wastes are contained in drums for offsite disposal (data from NPDES file).

In May, 1981, the Biology Section performed static acute toxicity bioassays on a sample of discharge from this facility. At that time, the sample was not acutely toxic to the organisms, Daphnia pulex, Simocephalus serrulatus, or Pimephales promelas (FDER 1981).

To again evaluate the extent of toxicity of this discharge to the biota of the receiving waters, the Southwest District office requested that additional bioassays be performed. Three separate 48 hour static acute toxicity bioassays were performed from 15 to 17 August, 1985. The results of these tests are presented in this report. Those involved in performing the tests and preparation of this report were: R. B. Frydenborg, M. L. Moultrie, and M. L. Roll.

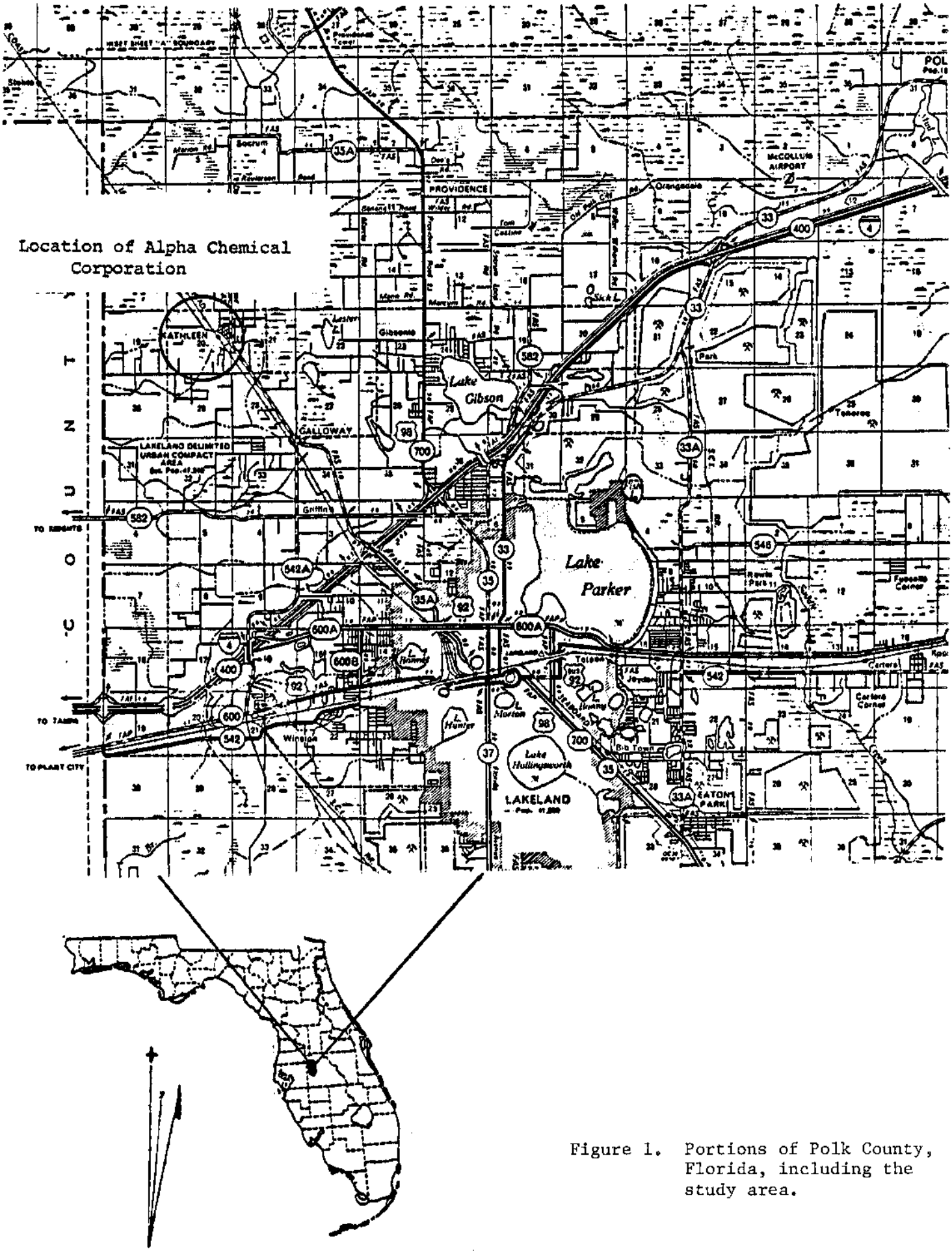


Figure 1. Portions of Polk County, Florida, including the study area.

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 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.

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 3 to 4 weeks old at the time of the test.

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

Test Methods

Methods used to perform acute toxicity bioassays are described by Peltier and Weber (1985) and the FDER Biology Section (1985). Some of the methods described below are more specific than those of Peltier and Weber. These will be noted in more detail.

Sampling Methods and Test Dates

Three separate 48 hour static acute toxicity bioassays were performed on a sample of effluent collected from the Alpha Chemical Corporation. The tests were performed from 1010 hours on 15 August to 0930 hours on 17 August, 1985.

The sample was collected by Ms. Nan Baggett of DER's Southwest District office at 1415 hours on 14 August, 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.

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 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, 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.

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 through 4.

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

Ceriodaphnia dubia (water flea)

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, Daphnia pulex, static acute toxicity bioassay of Alpha Chemical Corporation, Polk County, Florida, NPDES #FL0029653 on 15 to 17 August, 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	10	8.5	9.0	9.0	7.6	7.9	7.9	21.5	19.4	21.1			Control Water
Control	10	10	10	8.5	9.0	9.0	7.6	7.9	7.9				210	220	NH_3(as NH_3)(mg/l) 0 — Total Alkalinity 98 (mg/l $CaCO_3$)
10.0%	10	10	10	8.4	9.0	8.9	7.6	7.9	7.9	21.8	19.4	20.8			
10.0%	10	10	10	8.4	9.0	8.8	7.6	7.9	7.9				210	220	
32.0%	10	10	10	8.4	9.0	8.7	7.7	8.0	7.9	21.5	19.3	20.8			Total ammonia (as N)(mg/l) 0 — Total Hardness 105 (mg/l $CaCO_3$)
32.0%	10	10	10	8.4	8.9	8.6	7.7	8.0	7.9				205	212	
56.0%	10	10	10	8.5	8.9	8.5	7.8	8.0	7.9	21.1	19.3	20.7			
56.0%	10	10	10	8.5	8.9	8.5	7.8	8.0	7.9				200	210	
100.0%	10	10	10	8.5	9.0	8.3	7.9	8.0	7.9	20.5	19.4	20.8			
100.0%	10	10	10	8.5	8.9	8.2	7.9	8.0	7.9				190	208	100% Effluent
															NH_3(as NH_3)(mg/l) 0 — Total Alkalinity 95 (mg/l $CaCO_3$)
															Total ammonia (as N)(mg/l) 0 — Total Hardness 108 (mg/l $CaCO_3$)

Table 3. Data recorded during 48 hour, Ceriodaphnia dubia, static acute tocicity bioassay of Alpha Chemical Corporation, Polk County, Florida, NPDES #FL0029653 on 15 to 17 August, 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	10	8.2	8.5	8.2	7.7	7.9	8.0	20.9	19.8	20.9		
Control	10	10	8	8.2	8.5	8.2	7.7	7.9	8.0				210	220
10.0%	10	9	7	8.3	8.9	8.4	7.7	8.0	8.0	21.7	19.3	20.6		
10.0%	10	9	6	8.3	8.9	8.5	7.7	8.8	8.0				210	218
32.0%	10	10	10	8.4	9.0	8.5	7.8	8.0	8.0	21.4	19.0	20.5		
32.0%	10	10	9	8.4	9.0	8.5	7.8	8.0	8.0				205	212
56.0%	10	10	10	8.4	9.0	8.5	7.8	8.9	7.9	21.0	18.9	20.5		
56.0%	10	10	10	8.5	9.1	8.5	7.8	8.0	7.9				200	210
100.0%	10	10	9	8.5	9.1	8.5	8.0	8.0	7.9	20.4	18.9	20.7		
100.0%	10	10	10	8.6	9.1	8.5	8.0	8.0	7.9				190	205

Control Water

NH_3(as NH_3)(mg/l) 0

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

Total Alkalinity 98 (mg/l $CaCO_3$)

Total Hardness 105 (mg/l $CaCO_3$)

100% Effluent

NH_3(as NH_3)(mg/l) 0

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

Total Alkalinity 95 (mg/l $CaCO_3$)

Total Hardness 100 (mg/l $CaCO_3$)

Table 4. Data recorded during 48 hour, Notropis leedsi, static acute toxicity bioassay of Alpha Chemical Corporation, Polk County, Florida, NPDES #FL0029653 on 15 to 17 August, 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	8.5	8.5	8.4	7.6	7.6	7.9	21.6	19.2	20.7		
Control	5	5	5	8.5	8.5	8.5	7.6	7.6	7.9				210	225
10.0%	5	5	5	8.5	8.5	8.5	7.7	7.6	7.9	21.6	19.0	20.6		
10.0%	5	5	5	8.5	8.5	8.5	7.7	7.6	7.9				210	220
32.0%	5	5	5	8.4	8.5	8.4	7.8	7.7	7.9	21.3	18.9	20.3		
32.0%	5	5	5	8.4	8.5	8.5	7.8	7.7	7.9				208	215
56.0%	5	5	5	8.5	8.6	8.4	7.9	7.7	7.9	21.0	18.8	20.3		
56.0%	5	5	5	8.6	8.6	8.4	7.9	7.7	7.9				200	212
100.0%	5	5	5	8.5	8.7	8.3	8.0	7.9	7.9	20.5	18.8	20.5		
100.0%	5	5	5	8.5	8.7	8.2	8.0	7.8	7.9				195	210

Control Water

NH_3(as NH_3)(mg/l) 0

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

Total Alkalinity 98 (mg/l $CaCO_3$)

Total Hardness 105 (mg/l $CaCO_3$)

100% Effluent

NH_3(as NH_3)(mg/l) 0

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

Total Alkalinity 95 (mg/l $CaCO_3$)

Total Hardness 108 (mg/l $CaCO_3$)

RESULTS AND DISCUSSION

The sample of effluent collected from the Alpha Chemical Corporation was not acutely toxic to the test organisms, Daphnia pulex, Ceriodaphnia dubia, or Notropis leedsi. No mortality occurred in the D. pulex and N. leedis 48 hour bioassays (Tables 2 and 4), and only random mortality was noted in the C. dubia 48 hour test (Table 3). Chemical analysis were not performed since the sample was not toxic.

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