

The Effects Of Selected Herbicides On Phytoplankton And Sulfur Bacteria Populations¹

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ABSTRACT

Effects of three salt formulations of endothall: Mono (dimethytridecylamine) oxide-7-oxabicyclo(2.2.1)heptane-2,3-dicarboxylic acid equivalent (TD-1874); Mono(N,N-dimethylakylamine)-7-oxabicyclo(2.2.1) heptane-2,3-dicarboxylic acid equivalent (Hydrothol 191); Dihydroxy aluminum salt of 7-oxabicyclo(2.2.1)heptane-2,3-dicarboxylic acid equivalent (System E); and 6.7-dihydrodipyrido(1,2-e; 2', 1'-c) pyrazinedium dibromide (Diquat) plus triethanolamine complex of copper sulfate (Cutrine Plus) on phytoplankton and bacteria populations, correlated with chemical, temperature and macrophyte growth data, were evaluated in test pools in central Florida. All chemicals provided complete control of the submersed macrophyte community. Pools treated with all chemicals except the dihydroxy aluminum salt of endothall were characterized by high turbidity resulting from dense sulfur bacteria and phytoplankton blooms and putrid odors of H₂S gas from decaying fish, invertebrates, and macrophytes. Pools treated with the dihydroxy aluminum salt of endothall were characterized by low turbidity, no unpleasant odor and abundant growth of muskgrass (*Chara* sp.).

INTRODUCTION

When aquatic macrophytes die and decompose, either naturally or by chemical treatment, natural phytoplankton populations are sometimes subjected to adverse macro and micro-nutrient levels during the biological decomposition of the organic matter.

Many chemical and physical factors affect the presence or absence, distribution and abundance of algae in a water system. Several factors which regulate and influence the growth of algae populations to nuisance levels (bloom) are: temperature, season, oxygen, pH, ortho-phosphates, total organic nitrogen, turbidity, sulfates, and alkalinity (8, 14).

Unpleasant odors during the microbial decomposition of aquatic plants result in part from the mineralization of organic sulfur compounds and subsequent liberation of sulfur in the reduced inorganic form (H₂S). Sulfur is available to living organisms principally in the form of soluble sulfates. These are recognized as a required nutrient for most plants, and are utilized in cellular metabolism in forms such as the sulphydral groups of the amino acid,

cystine. Eventually, these organic sulfur compounds are utilized as nutrients by microorganisms that decompose plants. During the decomposition of the organic residues by microbes, the sulfur present in organic combination is changed, after several transformations, into inorganic forms such as hydrogen sulfide (under anaerobic conditions) and sulfate (under aerobic conditions). Certain microbes which either oxidize or reduce significant amounts of inorganic sulfur compounds are the sulfur bacteria (3, 16, 17).

The purpose of this study was to monitor the changes in phytoplankton and sulfur bacteria populations over a 9-month period (August 1974 to May 1975) after application of the following herbicides: TD-1874; Hydrothol 191; System E; and Diquat plus Cutrine Plus. Data were correlated to reflect changes in frequency of occurrence, variation in percent total population, relative abundance of dominant genera, and generic succession, following treatments with various herbicides.

METHODS AND MATERIALS

This study was part of an overall investigation on the seasonal fluctuation of some aquatic plants; and the subsequent effects on the biota and water quality following treatment with selected herbicides.²

Forty-five plastic pools (0.91 m in depth by 3.66 m in diameter) were filled with washed sand to a depth of 15 cm and planted with the following aquatic macrophytes in various combinations: hydrilla (*Hydrilla verticillata* Royle); Eurasian watermilfoil (*Myriophyllum spicatum* L.); coontail (*Ceratophyllum demersum* L.); muskgrass; eelgrass (*Vallisneria americana* Michx.); and southern naiad (*Najas quadalupensis* (Sprengel) Magnus). Water levels were stabilized and continuously maintained through an irrigation system connected to the Eustis city water supply. These plants were allowed to establish for approximately 1 year. Representatives from the different microcosms were then selected at random for the various herbicide treatments and controls. Applications of the four herbicides were administered at the following rates: TD-1874,³ 3 mg per liter, replicated nine times; Hydrothol 191, 3 mg per liter, replicated seven times; System E, 0.93 kg/ha, (3.3 mg per

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²Hestand, R. S. and C. C. Carter. 1975. Ecology of aquatic plants. Florida Department of Natural Resources, Bureau of Aquatic Plant Research and Contr. pp. 125.

³A coded experimental compound made by Pennwalt Corp.

liter), replicated nine times; Diquat plus Cutrine Plus, 1 mg per liter and 0.33 mg per liter, respectively, replicated nine times; and the control replicated nine times. From these, two were selected from each group for extensive analysis. All determinations were conducted by accepted standard techniques (18).

Samples were collected concurrently with chemical, temperature, and growth-rate data. Chemical analysis of ortho-phosphates, total organic nitrogen, turbidity and pH were taken prior to treatment from all pools and a mean determined to established a baseline. Experimental data were then plotted as deviation from the baseline (positive or negative) to reflect changes in water quality. Actual readings are given throughout the text (Figures 1 and 2). Samples were taken every 2 days during the first 4 weeks (to monitor initial treatment effects), weekly for the following 42 days, and monthly thereafter.

From each station a 200-ml sample was taken with a 2-liter Kemmerer sampler at a depth of 0.31 m. Within 72 hr algae were prepared for identification as follows: a 40-ml aliquot of the sample was centrifuged for 5 minutes at 1840 X G, the supernate poured off, and more water from the sample was added and centrifuged. This was repeated until the entire 200-ml sample was reduced to a 5-ml concentrate. Algae were then resuspended and preserved in 10 ml of a stock solution. This was prepared by mixing 4 ml of 40% formaldehyde; 5 ml of 20% solution of clear detergent; and 1 ml of a saturated cupric sulfate solution; and distilled H₂O was added to obtain a standard volume of 100 ml.

Several taxonomic keys were used to identify algae and bacteria (12, 13, 15, 5, 18, 1). For quantitative analysis a 0.10-ml sample was removed from the 15-ml concentrate and placed in a Palmer counting cell. Organisms in 10 randomly

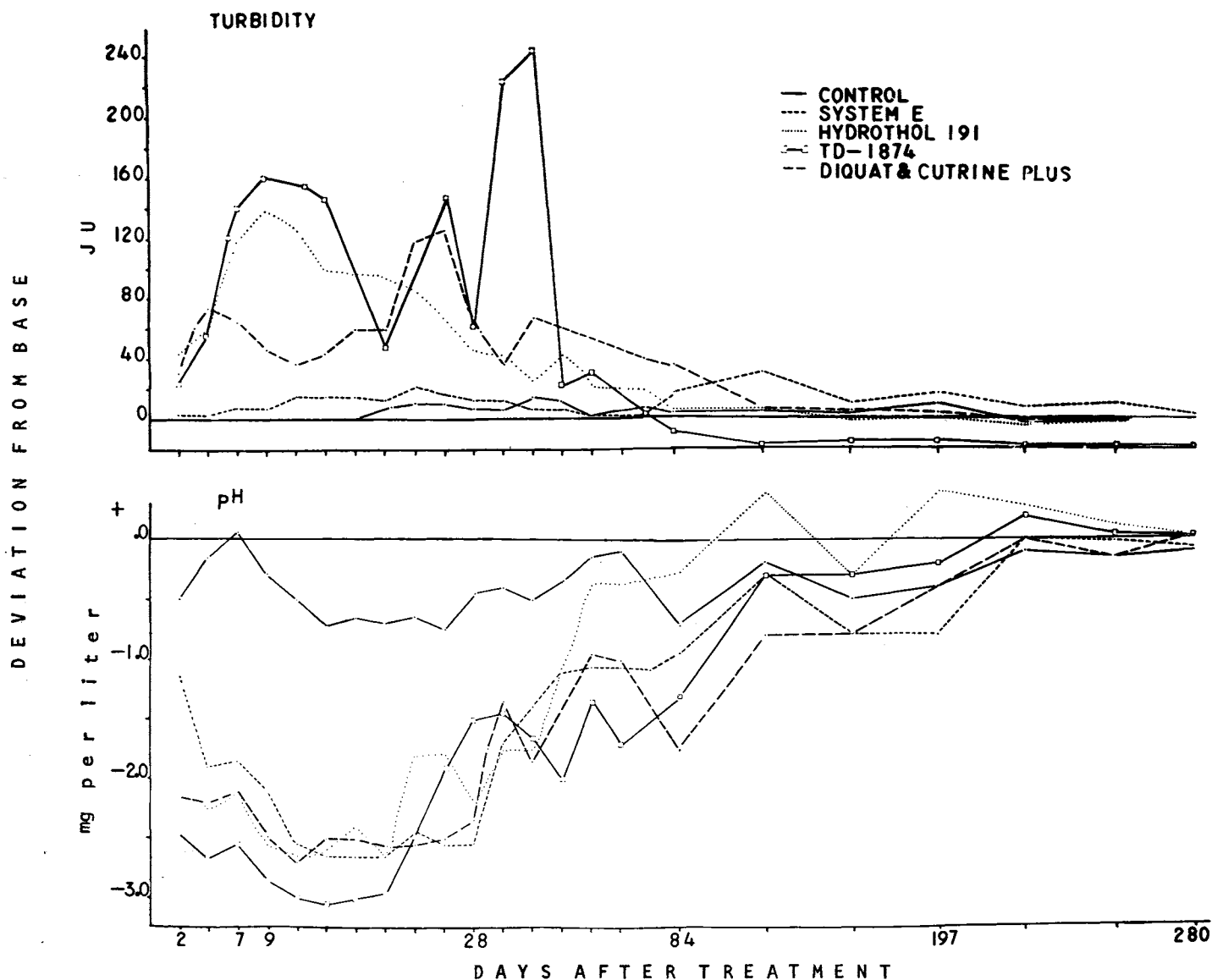


Figure 1. Comparison of Water Quality in Treated and Untreated Pools for Analysis of Turbidity and pH.

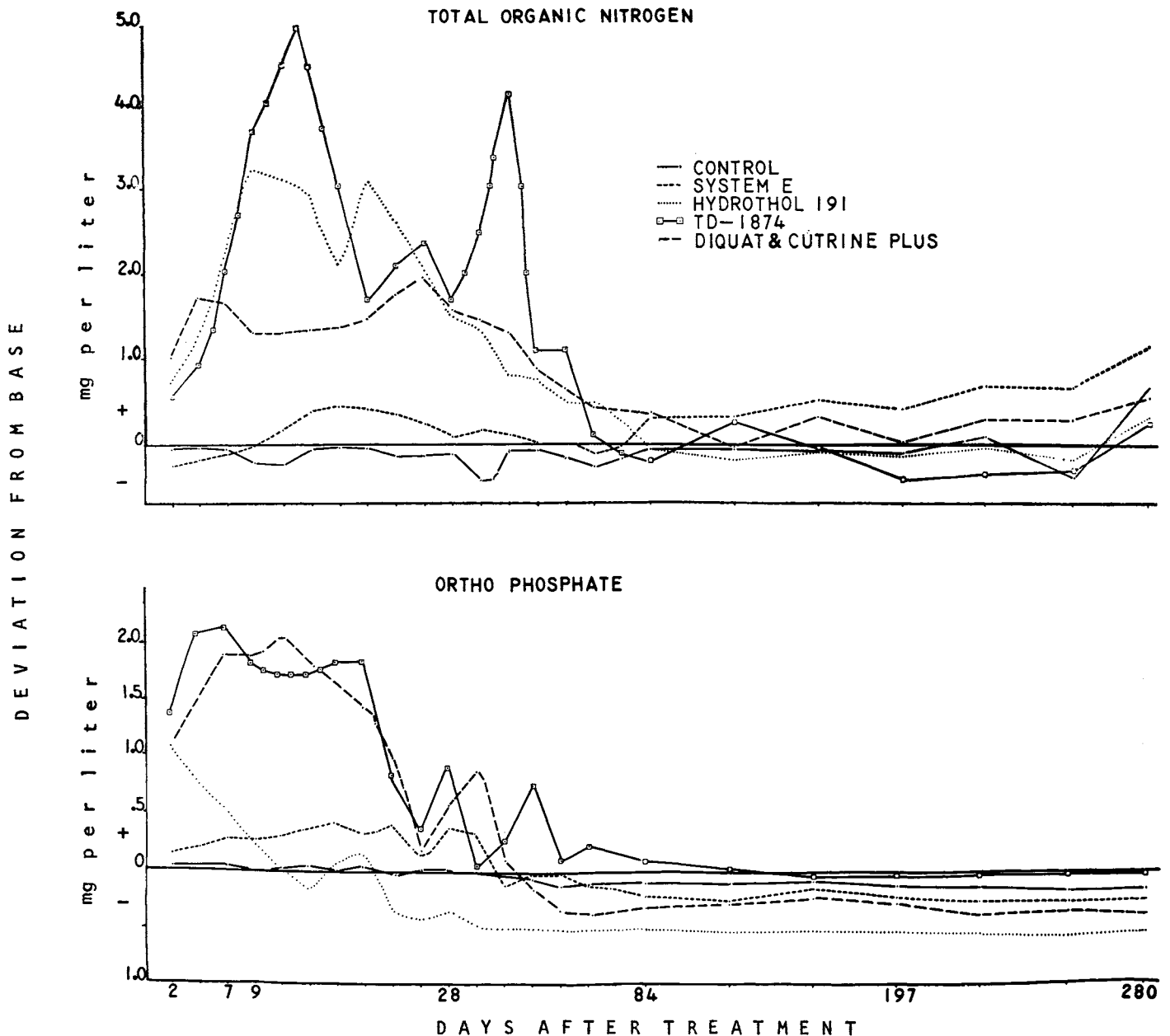


Figure 2. Comparison of Water Quality in Treated and Untreated Pools for Analysis of Total Organic Nitrogen and Ortho-Phosphates.

selected fields on hi-power (430X) were then counted, the average number of each type per field determined and expanded to number per liter.⁴

RESULTS AND DISCUSSION

Sulfur bacteria are comprised of three groups, two of which were observed in the pools. Representing the green and purple (or red) group were *Thiopedia* (nearly spherical, non motile), *Chromatium* (motile, short rods, kidney shaped), *Thiospirillum* (motile, spiral cells), and *Chlorobium* (green-sulfur bacteria, non motile, oval). Most

organisms in this group are relatively large, packed with sulfur globules, and pigmented to the point where even single individuals appear reddish or faint yellowish-green in color. These can be recognized visually when they occur in large quantities. Group two comprised of colorless, filamentous bacteria, was represented by *Beggiatoa* (1, 18).

Control Pools. Water in the control pools remained clear throughout the entire study. Aquatic macrophytes present remained in a stable and healthy condition, though several transitions of succession did occur (Footnote 2). Water chemistry data indicated that the turbidity averaged 11 JU (Jackson Units), well within the "recommended limit" of 5 JU and the "acceptable limit" of 25 JU set by the World Health Organization for drinking water standards (19). The pH values also remained within the

⁴Wilbur, Robert L. 1971. Habitat Manipulation. Annual Report. Florida Game and Fresh Water Fish Commission, D-J Project F-26-2, (Limnological Sampling). pp. 1-3.

required 6.5 to 9.2 limit at 9.0. Ortho-phosphates never exceeded 0.39 mg per liter and total organic nitrogen averaged 0.77 mg per liter (Figures 1 and 2). Hydrosoil interface water temperatures ranged from 22.2 to 27.8 C for the first 63 days, then dropped to 13.6 C on 13 December, 113 days after herbicide treatments. These low readings continued through March, 217 days after treatment (dat), when temperatures increased to the 22.2 C to 25.0 C range.

Generic succession within the algal community was variable. Differences among samples in total number of organisms were minor and could be attributed to normal seasonal fluctuations (Figure 3 and Tables 1-4). Six major green algae genera were found, representing five families in four orders. *Selenastrum minutum* was the only species found in quantity (70.4×10^6 per liter) and populations appeared to peak as interface water temperature declined. This algae group is noted for high photosynthetic rates in cooler waters (2). *Selenastrum* is a very small algae, how-

ever, and is only 2 to 3 μ in diameter and 7 to 9 μ between apices. Lackey, 9, arbitrarily set 0.5×10^6 per liter as a defined bloom but did stipulate that large numbers of extremely small algae, as *Gloeocapsa* (blue-green), though quantitatively considered a bloom, show no evidence of water discoloration nor nuisance problems in some instances. Blue-green algae were represented by only two genera, *Anacystis* and *Oscillatoria*, with *Anacystis* the dominant of the two. These are found in order *Chroococcales* and order *Hormogonales*, respectively. Six genera dominated the yellow-green group. They are distributed within four families, all in the order *Pennales*. The flagellate group had only one genus, *Euglena*, which was recorded sporadically. The last three groups had relatively low quantitative readings throughout the study. Percent frequency of occurrence indicated that the green algae occurred 100% of the time; blue-green, 67%; yellow-green, 54%; and flagellates, 42%. No bacteria were recorded in

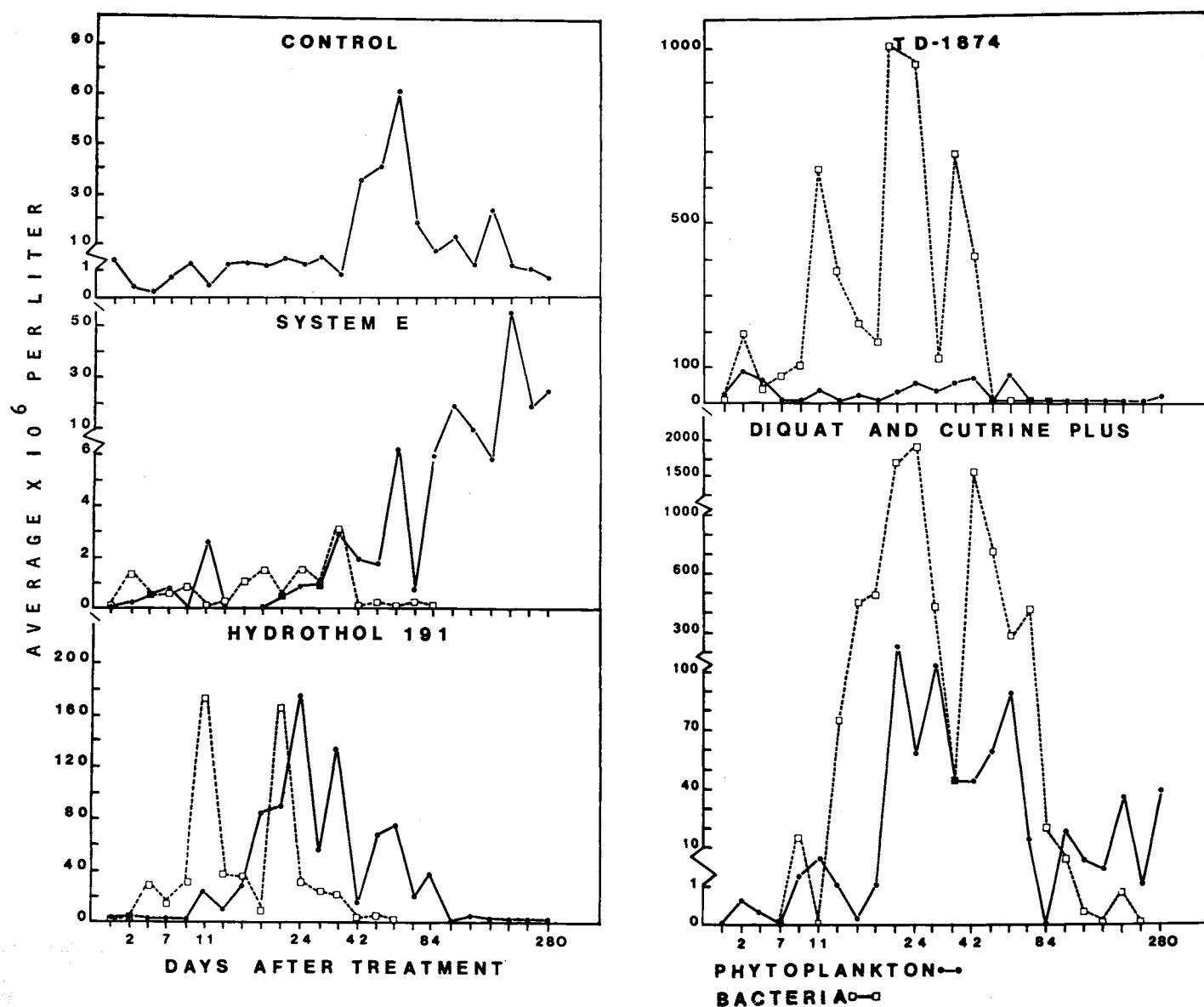


Figure 3. Effects of Various Herbicide Treatments on Phytoplankton and Bacteria Populations.

TABLE I. DOMINATE GREEN PHYTOPLANKTON (CHLOROPHYTA) SUCCESSION IN POOLS AFTER APPLICATION OF VARIOUS HERBICIDES.

Days After Treatment	Herbicide Treatment				
	Control	Hydrothol 191	System E	TD 1874	Diquat Plus Cutrine
0	<i>Staurastrum</i>	<i>Staurastrum</i>		<i>Gloeocystis</i>	<i>Gloeocystis</i>
2	<i>Ulothrix</i>	<i>Spirogyra</i>			
4	<i>Staurastrum</i>		<i>Chlamydomonas</i>		
7			<i>Cosmarium</i>		
9	<i>Gloeocystis</i>	<i>Gloeocystis</i>			
14					<i>Chlorococcum</i>
16				<i>Chlamydomonas</i>	
18		<i>Chlamydomonas</i>			<i>Scenedesmus</i>
22			<i>Tetraedron</i>		<i>Selenastrum</i>
24		<i>Gloeocystis</i>	<i>Gloeocystis</i>	<i>Selenastrum</i>	
28				<i>Chlamydomonas</i>	
35		<i>Selenastrum</i>	<i>Selenastrum</i>	<i>Scenedesmus</i>	<i>Gloeocystis</i>
42	<i>Selenastrum</i>	<i>Gloeocystis</i>		<i>Selenastrum</i>	<i>Chlamydomonas</i>
49		<i>Coelastrum</i>	<i>Coelastrum</i>	<i>Scenedesmus</i>	
56			<i>Gloeocystis</i>	<i>Gloeocystis</i>	
63				<i>Ankistrodesmus</i>	
84				<i>Dictyosporium</i>	
113	<i>Cosmarium</i>		<i>Selenastrum</i>	<i>Chlamydomonas</i>	<i>Selenastrum</i>
156			<i>Cosmarium</i>		<i>Gloeocystis</i>
197	<i>Ankistrodesmus</i>	<i>Gloeocystis</i>		<i>Gloeocystis</i>	
217	<i>Cosmarium</i>	<i>Scenedesmus</i>		<i>Chlamydomonas</i>	<i>Selenastrum</i>
245		<i>Tetraedron</i>		<i>Gloeocystis</i>	<i>Gloeocystis</i>
280	<i>Gloeocystis</i>	<i>Chlamydomonas</i>		<i>Selenastrum</i>	<i>Selenastrum</i>

the control pools. By total numbers, green algae represented 91% of the population; blue-green, 6%; flagellates, 3%; and yellow-green, 1% (Table 5).

Hydrothol 191. Within 2 dat, plants in pools treated with Hydrothol 191 were turning brown and starting to die with the water dark tannin stained in appearance. Turbidity increased as high as 60 JU, ortho-phosphate and total organic nitrogen increased to 1.65 mg per liter and 1.60 mg per liter, respectively, and the pH dropped from 9.4 to 7.4 (Figures 1 and 2). Phytoplankton exhibited no quantitative changes, while the aerobic bacteria *Beggiatoa*, was found in trace amounts (Figure 3, Table 6).

All plants appeared dead and several were decomposing 9 dat with the Hydrothol 191 treatment. Hydrogen sulfide (H₂S) liberation was noted as the pH dropped to 6.9. High bacteria counts were recorded in the form of the anaerobic red sulfur bacteria *Thiopedia*, at 31.9 X 10⁶ per liter (Figure 3, Table 6). The water turned red, and a reddish brown scum was evident at the surface. Turbidity had

increased to an average of 144 JU, ortho-phosphate fluctuated slightly and started to decline, and total organic nitrogen increased to an average of 4.0 mg per liter. Phytoplankton populations exhibited no quantitative changes, though several generic changes had occurred.

By the end of 22 days, high counts (165 X 10⁶ per liter) of the green anaerobic bacteria *Chlorobium*, were recorded, with the water turning green. A slight increase in the pH and indications of plant regrowth were noted (Footnote 2). Ortho-phosphates decreased to normal concentrations as a moderate phytoplankton bloom occurred in the form of the blue-green algae *Anacystis* and *Oscillatoria*, at 39.7 X 10⁶ per liter and the flagellate Euglenoid at 81.4 X 10⁶ per liter (Tables 1, 2, and 4, Figure 3).

On 7 October, 56 dat, the water was fairly clear to greenish in color with moderate phytoplankton counts dominated by green algae, *Coelastrum* (Table 1). Turbidity, ortho-phosphate, and pH had returned to normal while total organic nitrogen readings dropped to 1.36 mg per

TABLE 2. DOMINATE BLUE-GREEN PHYTOPLANKTON (CYANOPHYTA) SUCCESSION IN POOLS AFTER APPLICATION OF VARIOUS HERBICIDES.

Days After Treatment	Herbicide Treatment				
	Control	Hydrothol 191	System E	TD 1874	Diquat Plus Cutrine
0				<i>Chroococcus</i>	
2		<i>Anabaena</i>		<i>Oscillatoria</i>	
4			<i>Anacystis</i>	<i>Gloeotrichia</i>	<i>Anacystis</i>
7	<i>Anacystis</i>	<i>Anacystis</i>		<i>Anacystis</i>	
9					
11	<i>Anacystis</i>	<i>Anabaena</i>		<i>Anabaena</i>	
14				<i>Anacystis</i>	
16		<i>Oscillatoria</i>			
18					
22		<i>Anacystis</i>	<i>Oscillatoria</i>	<i>Oscillatoria</i>	<i>Oscillatoria</i>
24		<i>Oscillatoria</i>	<i>Anacystis</i>		
28				<i>Anacystis</i>	<i>Anacystis</i>
35					
49	<i>Oscillatoria</i>	<i>Anacystis</i>			<i>Anacystis</i>
56					
63					
84					
113					
156	<i>Anacystis</i>	<i>Anacystis</i>			
197					
217					
245					
280			<i>Spirulina</i>		

liter (Figures 1 and 2). Sulfur bacteria decreased to 3.8×10^6 per liter 49 dat with *Chlorobium* still dominant. By 7 October, bacteria populations disappeared (Table 6).

By 4 November, 84 dat, the microcosm stabilized with the reoccurrence of higher aquatic plants. Water quality and phytoplankton populations fluctuated in a similar manner to the control. All phyla were well represented during the study except the yellow-green algae (Tables 3 and 5). Percent frequency of occurrence indicated the sulfur bacteria were found 58% of the time with a recorded high count of 171.5×10^6 per liter before subsiding.

System E. Several plants had started to die within 2 days in pools treated with System E; however, the water remained clear.

Within 24 days all plants were decomposing except muskgrass, which increased its dominance, and by 4 November (84 dat) muskgrass dominated up to 40 to 50% of the pool area, as several vascular species returned in low percentages (Footnote 2).

Through the duration of the study, water quality and phytoplankton populations closely followed those of the control. However, turbidity increased slightly, as the pH indicated a decrease during decomposition (Figures 1 and 2).

Sulfur bacteria were found 54% of the time but only in trace amounts (3.1×10^6 per liter). Bacteria succession from aerobic to anaerobic and back to aerobic conditions was similar to Hydrothol 191. All bacteria counts had subsided by 63 dat (Tables 5 and 6).

Phytoplankton populations were low, but several generic changes did occur (Figure 3, Tables 1-4). Percent frequency of occurrence indicated that green algae occurred 75% of the time; blue-green, 71%; yellow-green, 46%; and flagellates, 17%. By total numbers, green algae represented 75% of the total population; blue-green, 19%; yellow-green, 4%; and flagellates, 2% (Table 5).

Muskgrass was not affected by the chemical application but rather persisted and crowded out the affected plants,

TABLE 3. DOMINATE YELLOW-GREEN PHYTOPLANKTON (CHRYSOPHYTA) SUCCESSION IN POOLS APPLICATION OF VARIOUS HERBICIDES.

Days After Treatment	Herbicide Treatment				
	Control	Hydrothol 191	System E	TD 1874	Diquat Plus Cutrine
0					
2	<i>Diatomella</i>		<i>Navicula</i>		<i>Navicula</i>
4				<i>Denticulia</i>	
7				<i>Diatomella</i>	
9					
11			<i>Navicula</i>		<i>Navicula</i>
14					
16					<i>Navicula</i>
22					
24	<i>Denticulia</i>		<i>Frustulia</i>		
28	<i>Opephora</i>		<i>Opephora</i>		
35					
42	<i>Diatomella</i>		<i>Frustulia</i>		
49	<i>Opephora</i>				
56	<i>Campylodiscus</i>			<i>Frustulia</i>	
63					<i>Stauroneis</i>
84				<i>Amphipleura</i>	
113			<i>Amphipleura</i>		<i>Coscinodiscus</i>
156	<i>Amphipleura</i>	<i>Frustulia</i>	<i>Frustulia</i>		<i>Opephora</i>
197	<i>Opephora</i>	<i>Opephora</i>			
217	<i>Fragilaria</i>		<i>Amphipleura</i>		
245					
280					<i>Frustulia</i>

utilizing the nutrients released during decomposition of the vascular plants. Muskgrass capacity for nutrient removal hinders the growth of planktonic algae (4), thus eliminating not only high bacteria counts as plants decompose, but phytoplankton populations as well. This results in exceptional water clarity during plant re-establishment, which is obviously beneficial unless muskgrass reaches detrimental proportions and reduces the utility of the water. Coverage reached 45 to 50% of the pool area before stabilization at which time natural fluctuations re-established as remaining nutrients were absorbed leading to regeneration of several other aquatic macrophyte species. *Cosmarium* (green algae) was recorded in moderate numbers as muskgrass's inhibitory influence upon the system lessened during the last three months of the study. Muskgrass still maintained 50 to 55% coverage at the study's termination.

TD-1874. In pretreatment analysis of pools treated with TD-1874, the water appeared light green, with the green

algae *Gloeocystis* dominant in a moderate bloom condition. Water chemistry was normal, though turbidity was high at 22 JU.

Within 2 dat, several plants were turning chlorotic and flaccid. The water was dark tannin stained, and turbidity, ortho-phosphate and total organic nitrogen exhibited significant increases. Phytoplankton populations increased, utilizing the nutrients released. The aerobic bacteria *Beggiatoa* was recorded with high counts of 119.0×10^6 per liter (Figures 1-3).

High bacteria counts of 748.0×10^6 per liter were recorded 11 dat in the form of the anaerobic red sulfur bacteria *Thiopedia*, resulting in reddish brown to dark red water. Hydrogen sulfide liberation was noted similar to that in Hydrothol 191 treatments as the pH dropped to 6.7. All plants were dead and decomposing. Turbidity recorded at 178 JU, and ortho-phosphate, and total organic nitrogen had increased significantly from 1.45 mg per liter

TABLE 4. DOMINATE FLAGELLATE SUCCESSION IN POOLS AFTER APPLICATIONS OF VARIOUS HERBICIDES.

Days After Treatment	Herbicide Treatment				
	Control	Hydrothol 191	System E	TD 1874	Diquat Plus Cutrine
0	<i>Euglena</i>				
2				<i>Euglena</i>	
7			<i>Euglena</i>		
9					<i>Euglena</i>
11	<i>Euglena</i>	<i>Euglenoids</i>	<i>Euglenoids</i>	<i>Euglenoids</i>	<i>Euglenoids</i>
14					
16					
18					<i>Euglenoids</i>
22				<i>Euglena</i>	<i>Eudorina</i>
24			<i>Euglenoids</i>		
28					<i>Euglena</i>
35			<i>Euglenoids</i>		
42					
49	<i>Euglena</i>				
63		<i>Volvox</i>		<i>Pandorina</i>	
84	<i>Euglena</i>	<i>Pandorina</i>			
113					
156		<i>Euglena</i>			
197					<i>Euglena</i>
217					
245					
280				<i>Euglena</i>	<i>Euglena</i>

to 1.80 mg per liter and 1.90 mg per liter to 6.15 mg per liter, respectively. Phytoplankton populations were decreasing at this time.

Within 24 dat high bacteria counts of $1,027 \times 10^6$ per liter were recorded in the form of the green sulfur bacteria *Chlorobium*, causing the water to change from red to brownish-green to green. Turbidity had decreased slightly to 168 JU; ortho-phosphate decreased from 1.9 mg per liter to an average of 0.45 mg per liter; total organic nitrogen decreased 50% to 3.5 mg per liter; and the pH increased from 6.8 to 7.7 (Figures 1-3). Phytoplankton analysis showed several generic transitions had occurred. *Chlamydomonas* still dominated the green algae group and had increased to 23.0×10^6 per liter (22 dat). This was subsequently replaced by *Selenastrum minutum*, which received a count of 47.0×10^6 per liter. This involved a change from order *Volvocales* to order *Chlorococcales*. *Anacystis* replaced *Anabaena* 14 dat and persisted until 22 dat, when *Oscillatoria* became dominant at 3.5×10^6 per liter. This involved a shift in genera between two orders

and three families. *Euglena* was found sporadically reaching 18.0×10^6 per liter by 24 dat. Representatives from the yellow-green group were not recorded (Tables 1-4).

By 42 dat the water was fairly clear to greenish in color though the turbidity, pH, and ortho-phosphate remained basically constant. Sulfur bacteria *Beggiatoa* and *Chlorobium* both persisted through this period, with a combined total of 405.9×10^6 per liter; *Chlorobium* was the dominant of the two. Phytoplankton populations also decreased during this period as several plants had started to re-establish (Figure 3, Table 6).

By 4 November, 84 dat, all bacteria counts had subsided with *Beggiatoa* being the last bacteria recorded (Table 6). Water chemistry parameters returned to normal and water clarity was clear to brownish in color, as percent plant coverage continued to increase. Several generic changes occurred through this period as phytoplankton populations continued to decrease (Figure 3, Tables 1-4).

By 25 February, 197 dat, the percent plant coverage was 60 to 70%, indicating reestablishment into the system (Footnote 2). The water remained clear with fluctuations

in physical characteristics following a pattern similar to the fluctuations in the control. Phytoplankton populations increased moderately after sulfur bacteria counts subsided and produced similar patterns of fluctuations through the study's duration as those of the control. Percent frequency of occurrence indicated that green algae occurred 100% of the time; blue-green, 92%; flagellates, 54%; yellow-green, 29%; and sulfur bacteria, 54%. By total numbers, green algae represented 81% of the total populations; blue-green, 6%; flagellates, 12%; and yellow-green, 1% (Table 5).

Diquat plus Cutrine Plus. In pools treated with Diquat plus Cutrine Plus all plants appeared dead and decomposing within 9 days, and the water appeared brownish in coloration. Turbidity, ortho-phosphate and total organic nitrogen fluctuated, as the pH decreased from 9.0 to 7.0. Phytoplankton populations exhibited a slight increase, while the aerobic bacteria *Beggiatoa* was found in moderate concentrations (Figures 1-3).

Bacteria counts of 70.0×10^6 per liter were recorded 14 dat in the form of the anaerobic red sulfur bacteria *Thiopedia*. Water turned reddish brown in coloration, and H_2S liberation was recorded. Turbidity, ortho-phosphate and total organic nitrogen continued to increase. Phytoplankton populations exhibited no increases at this time, though several changes in genera occurred (Tables 1-4).

The water had turned green in color 24 dat, and bacteria were found in the form of the green sulfur bacteria *Chlorobium*. Turbidity was recorded at 135 JU, and ortho-phosphate had decreased. A heavy, mixed phytoplankton bloom occurred, consisting of representatives within the green genera *Selenastrum* and blue-green genera *Oscillatoria* at 233.0×10^6 per liter (Figure 3 and Tables 1 and 2).

By the end of 42 days high bacteria concentrations of *Chlorobium* were recorded ($1,646 \times 10^6$ per liter). The water appeared a dark brownish green and indications of plant regrowth were noted as total organic nitrogen decreased. Phytoplankton populations decreased, with several generic changes occurring (Figure 3).

Bacteria were rapidly decreasing 84 dat with both *Chlorobium* and *Beggiatoa* recorded (21.1×10^6 per liter).

The water was greenish in color and several plants were recorded. These appeared unhealthy and later died (Footnote 2). Phytoplankton populations still exhibited a marked decrease; dominated by the green algae *Chlamydomonas* and the blue-green genus *Anacystis*. Turbidity of 44 JU was recorded.

Not until 17 March, 217 dat, did this microcosm indicate stabilization of trends. Turbidity readings dropped steadily from 44 JU to 9 JU through this 133-day period, though water clarity was still a light green. Several aquatic macrophytes were well established by this sample period as ortho-phosphate, total organic nitrogen, and pH readings became comparable to those of the control. Phytoplankton populations also increased moderately as sulfur bacteria diminished. *Beggiatoa* (sulfur bacteria) recorded relatively low counts (1.0×10^8 per liter) suggesting its apparent and eventual demise as bacteria readings were last recorded 217 dat (Table 6). Water quality and phytoplankton populations thereafter exhibited fluctuations similar to the control as increasing plant coverage stabilized the microcosm.

While bacteria counts fluctuated erratically, as plants died and decomposed, a general trend in sulfur bacteria succession was observed in all the treated pools. First recorded was the aerobic bacteria *Beggiatoa* which usually persisted until the pH dropped below 8.0 (10). As the pH continued to drop, anaerobic bacteria proceeded to dominate and liberate H_2S by-products. Representatives from the red or purple group then emerged, as either *Thiopedia*, *Thiospirillum*, or *Chromatium*. These were followed (third in succession) by *Chlorobium* from the green sulfur group. Aerobic conditions returned as plants reestablished; *Beggiatoa* was recorded once again and persisted until the microcosm stabilized and sulfur related compounds were utilized by the plants.

Interface water temperatures were generally lower during the early stages of plant reestablishment. This correlated with the fact that green algae, diatoms, and dinoflagellates have high photosynthetic rates in cooler waters, and also obtain their maximum concentration in spring, or early summer, and in fall. Blue-green algae are adapted best to warmer temperatures, as recorded at the

TABLE 5. PERCENT FREQUENCY OF OCCURRENCE (FO), PERCENT TOTAL POPULATION (TP), AND RECORDED HIGH COUNTS (HC) OF PHYTOPLANKTON AND SULFUR BACTERIA IN PLASTIC POOLS WITH VARIOUS HERBICIDES.

Herbicide Treatment	Occurrence Condition	Green (%)	Phytoplankton		Flagellates (%)	Sulfur Bacteria (%)	HC Phyla ($\times 10^6$)	HC Bacteria ($\times 10^6$)
			Blue-green (%)	Yellow-green (%)				
Control	FO	100	67	54	42	—	71.4	—
	TP	91	6	1	3			
System E	FO	75	71	46	17	54	55.4	3.1
	TP	75	19	4	2			
Hydrothol 191	FO	79	75	17	63	58	173.3	171.5
	TP	37	29	1	33			
TD-1874	FO	100	92	29	54	54	99.6	1027.3
	TP	81	6	1	12			
Diquat Plus Cutrine Plus	FO	79	71	33	54	67	236.7	1861.7
	TP	92	4	1	3			

TABLE 6. BACTERIA SUCCESSION IN RELATION TO TREATMENT OF SUBMERSED MACROPHYTES WITH VARIOUS HERBICIDES.

Herbicide Treatment	Bacteria and Day Observed After Treatment												Total Number Of Days Observed
	Bacteria	Days	Bacteria	Days	Bacteria	Days	Bacteria	Days	Bacteria	Days	Bacteria	Days	
Control	---	---	---	---	---	---	---	---	---	---	---	---	---
System E	<i>Beggiatoa</i>	2	<i>Chlorobium Beggiatoa</i>	16	<i>Chromatium</i>	28	<i>Chlorobium</i>	35	<i>Beggiatoa</i>	49	<i>Beggiatoa</i> END	63	61
Diquat & Cutrine	<i>Beggiatoa</i>	9	<i>Thiopedia</i>	14	<i>Chlorobium Chromatium</i>	16	<i>Chlorobium</i>	24	<i>Chlorobium Beggiatoa</i>	83	<i>Beggiatoa</i> END	217	208
Hydrothol 191	<i>Beggiatoa</i>	2	<i>Thiopedia</i>	7	<i>Chlorobium Chromatium</i>	18	<i>Chlorobium</i>	22	<i>Chlorobium</i> END	49			47
TD-1874	<i>Beggiatoa</i>	2	<i>Thiopedia Thiosprillum</i>	9	<i>Chlorobium</i>	16	<i>Chlorobium Chromatium</i>	18	<i>Chlorobium Beggiatoa</i>	42	<i>Beggiatoa</i> END	63	61

beginning and end of the study. Alkaline waters produce fewer species, but in greater numbers, as observed after the microcosms had stabilized and phytoplankton populations fluctuated in apparent seasonal cycles (2, 6, 7, 14). The chemical control of large quantities of submerged aquatics can lead to increased levels of nitrogen and phosphorus in the water column and favor phytoplankton blooms (11).

In summary, the liquid herbicides caused a fast kill of the plants, resulting in a faster release of nutrients, which triggered an explosive growth rate in phytoplankton and bacteria populations. System E was the least detrimental of all the herbicides tested, due to the lack of these undesirable side effects.

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