CAUSAL FACTORS OF POPULATION VARIATION IN THE ALGAE OF A PAPERMILL WASTE WATER POND SYSTEM

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I. INTRODUCTION

Little information is available on the plankton organisms found in papermill waste impoundments, nor has there been correlation of the microflora with pond conditions. The present study was made to evaluate the effects of conditions in such an impoundment on algal populations during the first five years of operation.

The effects of two variations in operations were investigated. The first was the introduction of a settling basin after the reservoir had been in use for two years. No differences between the algal populations in the reservoir before and after construction of the settling basin were observed.

The second variation dealt with the manner in which the effluent was released from the impoundment. For the first two years of operation effluent was released from the bottom of the reservoir. During the third year of operation the effluent was released alternately from the bottom and from the top of the reservoir over a spillway. In the final two years of the study the water was released from the top. These operations resulted in water of different quality, with different algal populations.

Another purpose of the study was to determine whether changes in the quality of the water downstream were accompanied by qualitative or quantitative changes in algal populations. The number of genera increased but total number of organisms decreased as the waste water passed downstream.

This study of the algal population was made from 1957 to 1961 at the Crossett Company, Crossett, Arkansas, an integrated forests products manufacturer. Its operational divisions included paper mills, sawmill, wood chemical plants, and forestry. Large amounts of water were required for use as a carrier of the raw materials, and as a vehicle for the removal of the waste products in the manufacturing processes. Water consumption exceeded 25,000,000 gallons daily. Source of water was a series of deep wells located about 10 miles southeast of the mill grounds. Average discharge from the system was 25,000,000 gallons per day at TP1 (Fig. 1). The average discharge at R-1 reservoir was 23,000,000 GPD. Variation was the rule here as water level control and surface runoff after rains, as well as rates of evaporation and ground percolation were not constant.

The Crossett Paper Mill uses the Kraft process, in which a modification of the sulfate process reduces wood to fibers by the addition of chemicals which selectively removes lignin and other impurities, and isolates and partially purifies the fibers. Waste waters from the Crossett plant were typical of those from Kraft mills (Van Horn, 1949).

Wood is a highly complex association which includes cellulose, lignin, and hemicellulose. Of these substances cellulose is the most resistant to chemical action. Wood chips are carried to digesters where the chemicals are added and heat applied as steam pressure until the desired degree of fiber separation is obtained. During cooking, the chemical reacts with the lignin in the wood to form soluble compounds which may be removed by washing. The pulp stock is then passed through a system of vacuum washers and screens to remove the chemicals, undigested portion,and small fibers. The natant wash waters pass through tanks where the settleable material is retained, and the supernatant solution is returned to the digesters for re-use. The waste water, known as weak black liquor, is piped through evaporators where it is concentrated to 50-55% total solids. Tall oil, a mixture of the sodium salts of fatty and resinous acids, is skimmed off to decrease foaming and for sale as a valuable by-product. The black liquor is then taken to the recovery furnace where the remaining water is evaporated. The residue decomposes to form volatile gases, carbon,and inorganic salts. The volatile gases are burned to provide steam for electrical generation. Sodium is recovered as sodium carbonate. The remaining portions are discharged into the sewer as non-recoverable.

Refined pulp is transferred to the paper machine for sheet formation by the dispersal of the fibers across a screen. Water is removed and the pulp is compressed into a felt by gravitation and by suction from underneath. Water removed in this step is termed white water, most of which is returned for re-use. White water makes up the major portion of papermill effluent.

Wastes from the distillation processes in the wood chemical division form a large part of the final effluent. Accidental losses occur at many points in the system and contribute to the final effluent. The effluent system extends 15 miles from the plant grounds along a natural stream bed to the Ouachita River (Fig. 1). Aeration by passage through the 14 miles of stream channel increases the rate of gas exchange.

Changes were made in the natural drainage beginning about 1938. An ox-bow lake was converted into a holding pond (Mossy Lake, Fig. 3) by the construction of coffer dams at points of overflow. A 200 yard channel, equipped with a weir and gates, was excavated to the river. These additions made it possible to control the level of the lake and to adjust the outflow to the river. This pond had a capacity of more than a billion gallons. At maximum level the average depth of the lake was six feet. The estimated holding time was 45 days.

In 1956-57 a second holding pond (R-1, Fig. 2) was constructed 10 miles upstream from the first pond. This pond had a capacity of 613,000,000 gallons with an estimated holding time of 30 days. Stone rip-rapping extended one-half mile below the spillway to increase water turbulence and aeration. A gate located at the bottom of the dam permitted control of water volume and rate of discharge.

In 1959 two settling basins each 600 yards long by 100 yards wide were constructed two miles above the new holding pond (Fig. 1 and 6). The basins were constructed so that the effluent could be easily diverted from one to the other to permit removal of sludge.

The treated sewage effluent of the city of Crossett with a population of 6,000, was received into the total effluent at a point midway between the settling basins and the upper reservoir.

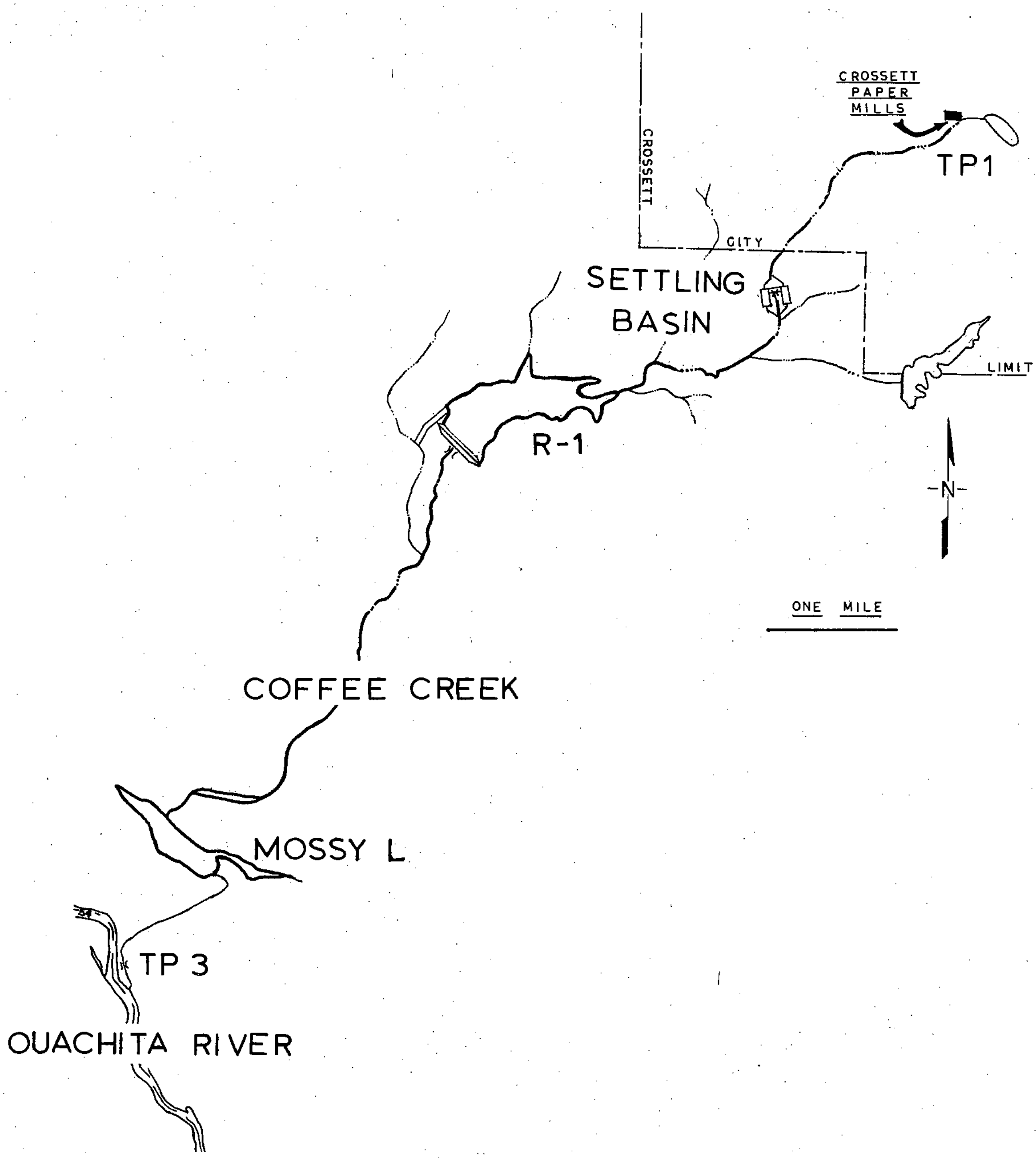

Fig. 1 Waste Disposal System. A portion of Coffee Creek has been omitted from this map.

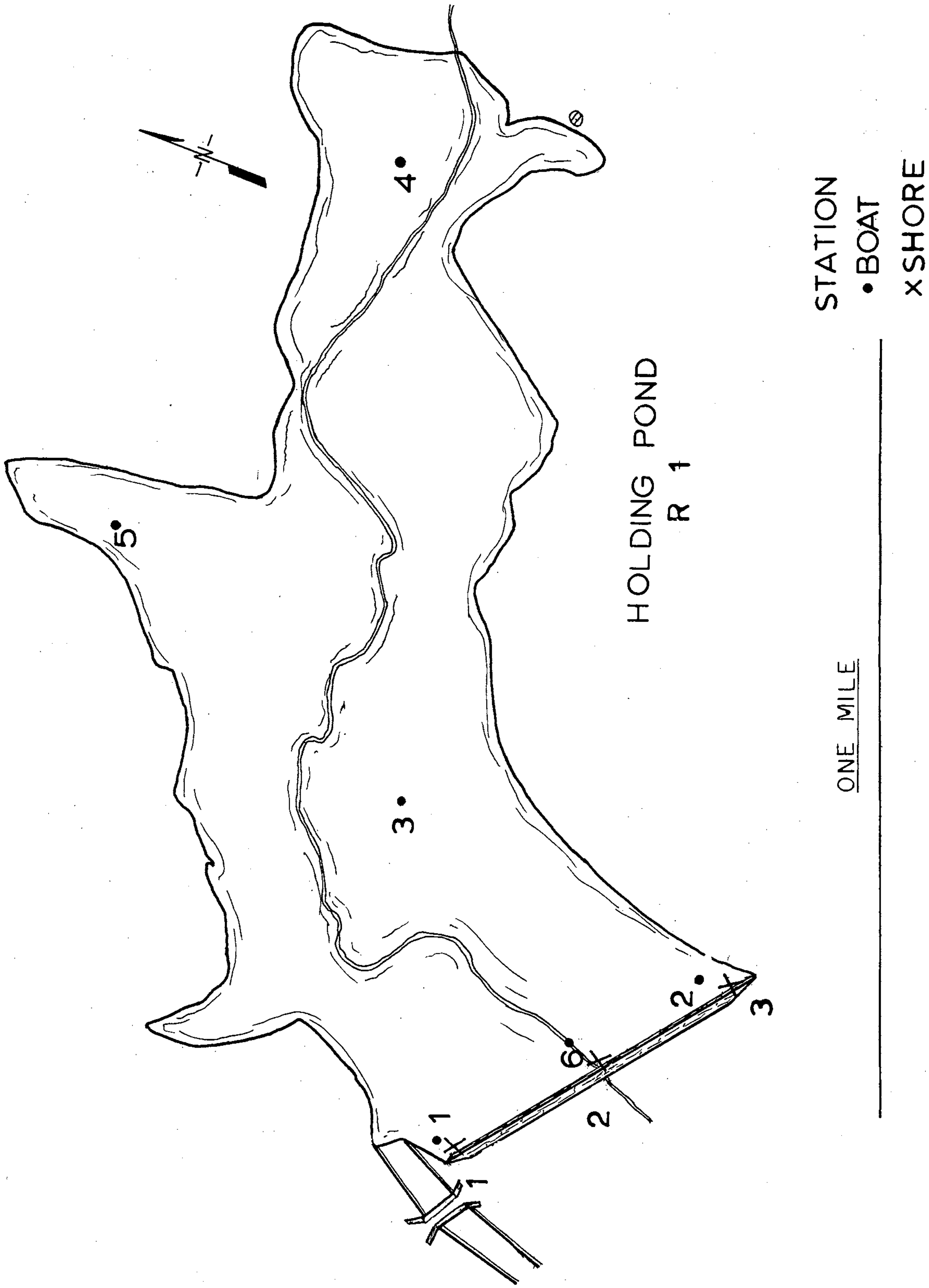

Fig. 2 Reservoir #1

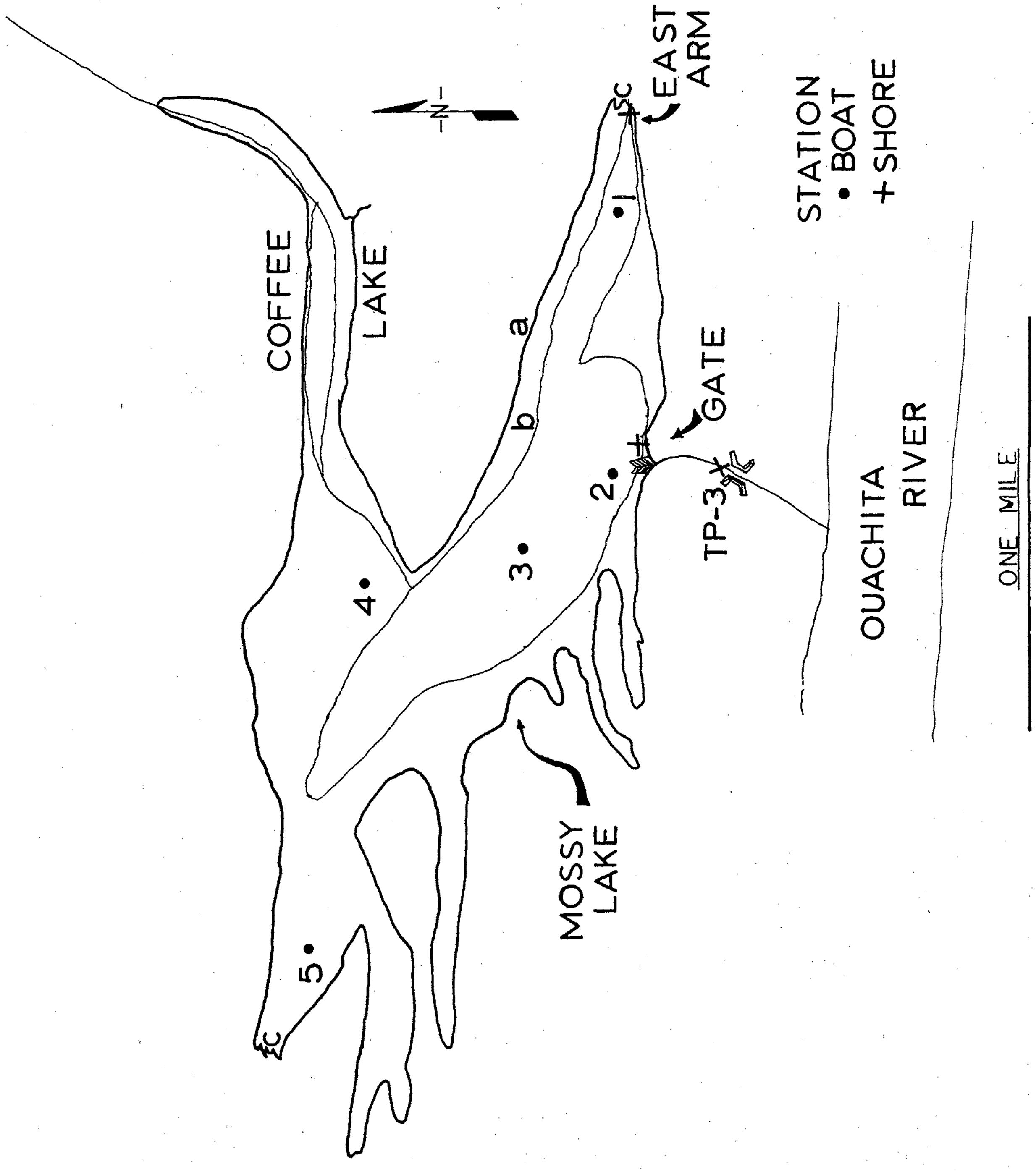

Fig. 3 Mossy Lake

a. enlarged lake margin
b. original lake margin
c. coffer dams

II. METHODS

Three sampling stations were established along the shores of each holding pond (Figs. 2 and 3). These stations were sampled three times a week for five years. In addition each of the holding ponds was divided into six areas with a sampling station centered in each. These stations were located at least ten feet from the shore and were sampled at widely separated time intervals from a boat.

All samples were collected with a triple-displacement sampler. Samples for physical and chemical determination were collected three feet below the water surface. A Beckman pocket model pH meter was used to determine pH. Dissolved oxygen was determined by the short Thireault modification of the Winkler method. Hydrogen sulfide content was estimated by a method of the carbon dioxide fluxing with lead acetate paper. Soda analysis was determined with a Beckman Flame Photometer. Five day biochemical oxygen demand (BOD) and chemical oxygen demand (COD) were determined according to methods described in Standard Methods of Water Analysis (APHA, 1955). Temperature was measured with a glass, mercury-filled centigrade thermometer. Light penetration values were obtained with a Secchi disk. Water color was measured against a platinum standard in a Beckman Spectronic 20 Colorimeter. Water flow was determined at a weir equipped with a Foxboro seven-day recorder. Water level was measured from a U. S. Geodetic Survey bench mark. Rainfall was recorded at a point near the papermills. Total solids were evaluated by evaporation of a 100 ml. aliquot.

Plankton samples were collected just below the water surface in the photosynthetic zone. Duplicate samples were taken at each sampling period at each of the collecting stations. Direct counts of the organisms were made of all samples reported in this study. Plankton samples were counted alive since presence of large quantities of debris made enumeration of preserved samples unreliable. Because of the large numbers involved, Chlorella was counted on a Spencer Brightline hemocytometer (Silva & Papenfus, 1953). All other planktonic organisms were counted in a Sedgewick-Rafter cell (Welch, 1940). Two counts were made of each sample. These data were arranged in a factorial design for orthogonal comparison (Steele and Torrey, 1960).

III. RESULTS AND CONCLUSIONS

A. Algal Succession in Papermill Waste

The genera of algae found in various parts of a waste treatment system depend on the tolerance of the algae to the waste products. Only the most tolerant algae will be found in the highly polluted areas. Less tolerant genera appear only after a certain degree of recovery has taken place. Progressive improvement downstream permits a succession of genera to appear. Algal succession may be interpreted by either of two methods. The presence of certain genera may be taken to denote the stage of recovery. In these cases the organism is called an indicator, and its presence is used as a gauge of the level of pollution. (Gaufin and Tarzwell, 1956). A second method of determining succession is to compare the relative number of genera and of individuals found at points in the system. This method also relies on the tolerance of the different algae to pollution. Less emphasis is placed on the individual and more attention is given to the relative composition of the community.

In this study, because of the seasonal variation in the kinds of genera found in the system, a single indicator could not be correlated with a definite stage in recovery at a given point; therefore, the second method of successional analysis has been employed. Introduction of algae that were not ordinarily present occurred many times as a result of runoff after a large rain. These algae did not become established because of their environmental requirements.

The number of genera increased as the effluent moved downstream (Fig. 4 and Table I). There was also an increase in the number of genera during the five years of the study. In surface samples from the upper reservoir the number of genera increased from 9 to 19 during the five year study period. At the same point, at the 10 foot depth, the number of genera increased from 7 to 11 in the first three years of the study but decreased to six in the last two years. This unusual difference may have been caused by change in the method of controlling the water level of the reservoir. Water was discharged alternately by release at the surface or 10 feet below the surface at the gate. The Chlorella population at the 10 foot level increased in total number at this time but populations of other genera decreased in number. The light penetration into this reservoir was only four inches when measured with a Secchi disk. Another factor was that the active algae were carried out of the reservoir by the removal of water at the surface before they had time to settle to a lower level.

The last sampling point was in the lower holding pond 50 yards above the receiving stream. There was a more diverse population of algae in this pond. The number of genera of algae increased from 16 in 1957 to 21 in 1960.

At two isolated locations in the lower reservoir a greater change in the number of genera represented was shown. In a bay on the east side of the lake the number of genera increased from 19 in 1957 to 25 in 1960. In a small basin formed at the exit from the channel, the number of genera increased from 21 in 1957 to 31 in 1960. Since these two points were somewhat isolated (Fig. 2), they had a lengthened holding time. These data indicate that increased holding time would yield

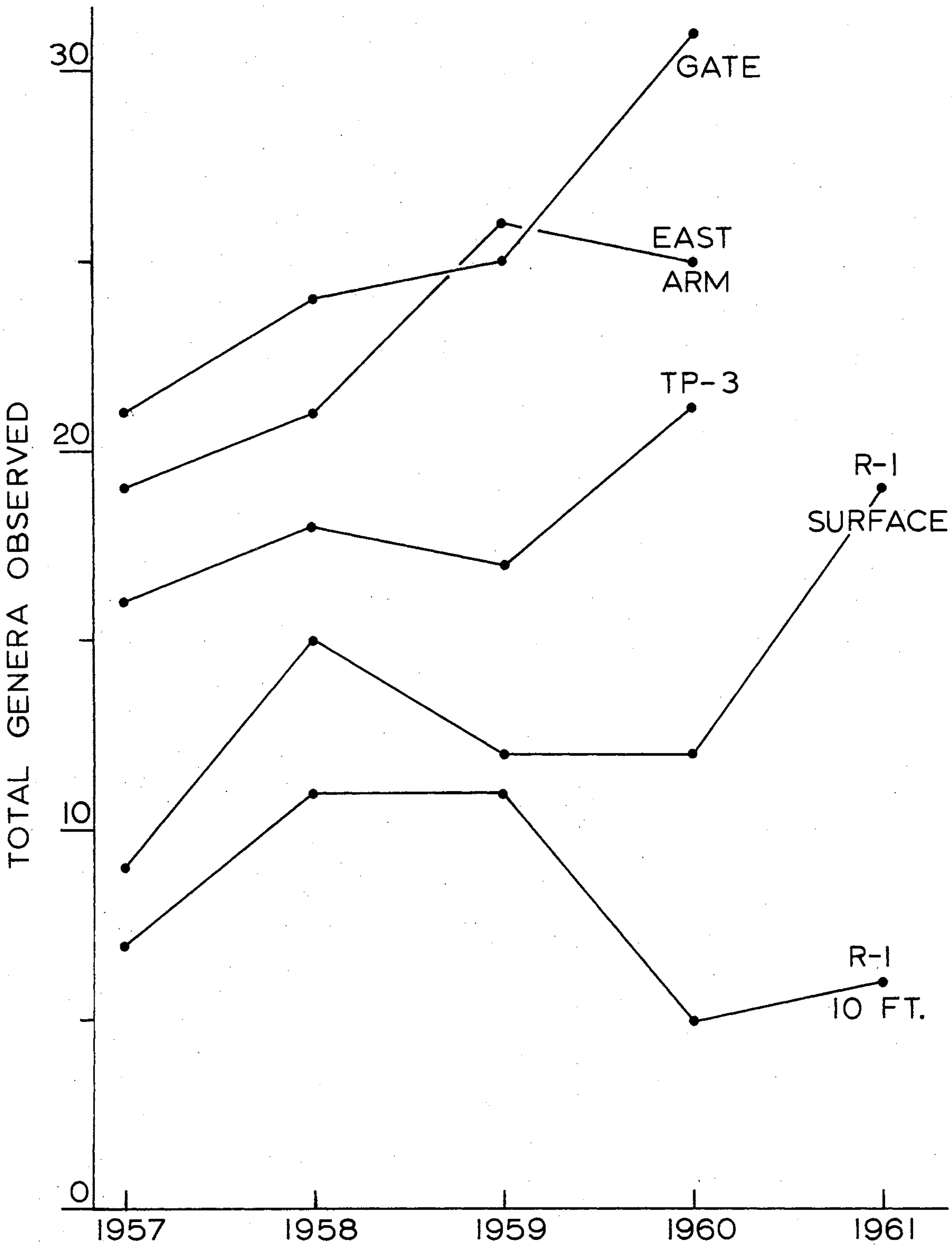

Fig. 4 Numeral Succession of Genera of Algae

TABLE I

ALGAL INCIDENCE IN PAPERMILL WASTE DISPOSAL SYSTEM

	Reservoir #1															Mossy Lake									
Station	**Weir Surface**					**Weir 10 Ft.**					**TP3**					**Gate**					**East Arm**				
Year	57	58	59	60	61	57	58	59	60	61	57	58	59	60	61	57	58	59	60	61	57	58	59	60	61
Chlorophyta																									
Chlorella	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Chlamydomonas	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Platydorina	X	X	X	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Chlamydobotrys	X	X	X	X	X	X	X	X	X		X	X				X	X		X	X	X	X	X	X	X
Pandorina	X			X	X						X	X		X		X	X	X	X	X	X	X	X	X	X
Eudorina	X	X	X		X		X	X			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Ankistrodesmus		X		X							X	X	X			X	X	X	X	X	X	X	X	X	X
Spirogyra			X										X	X			X	X		X					
Chlorococcum				X								X		X	X				X			X	X		
Volvox													X						X			X	X		
Actinastrum							X					X						X						X	X
Synura					X														X					X	
Pediastrum		X														X			X		X				
Cosmarium		X												X		X			X		X				
Scenedesmus	X																								
Tetraedron											X														
Ulothrix											X							X	X		X		X		
Schroederia																		X							
Euglenophyta																									
Euglena	X	X	X	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Phacus	X	X	X	X	X	X	X	X			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Trachelomonas					X																			X	
Chrysophyta																									
Navicula	X	X	X	X	X		X				X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Chrysococcus			X	X	X			X			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Synedra											X	X	X			X	X	X	X		X	X	X	X	
Fragillaria														X			X	X	X		X	X	X	X	
Cymbella					X																		X		
Uroglenopsis											X														
Pyrrophyta																									
Peridinium																	X								
Myxophyta																									
Anabaena		X													X	X			X	X			X	X	
Oscillatoria											X	X	X			X	X	X	X	X	X	X	X	X	X
Gleocapsa				X														X						X	
Lyngbya													X												
Microcystis											X														

a better quality of effluent.

Chlorella was the dominant genus of alga in the entire system. Next in order of importance were *Chlamydomonas*, *Euglena*, *Platydorina*, *Phacus*, and *Chlamydobotrys*. Oswald (1957) found *Chlorella* to be the dominant algae in zones of highest pollution and *Scenedesmus* as the dominant algae in zones of lowest pollution in sewage-oxidation ponds. Minter (unpublished data) found *Chlorella* to be the dominant algae in oil refinery oxidation ponds with other algae appearing in the late stages of stabilization.

Incidence of zooplankton was dependent on quality of the water (Table 2). *Paramecium* was the most common protozoan and was found at all points in the system. Other ciliates became common in the second reservoir. Rotifers were found at all points in the system. Rotifers found in the upper reservoir were associated with drifting mats of pulp (observe mats of pulp in Fig. 6). Copepods and *Daphnia* were found in the lower basin only.

Succession has been shown in this system when the variation in number of genera was used as a criterion for succession. Water with the heaviest pollution load had the smaller number of genera but the number of individuals was greatest. The mere presence of a particular organism does not make it a satisfactory indicator, since most organisms can be introduced as transients.

TABLE II

INCIDENCE OF ZOOPKLANKTON IN PAPERMILL WASTE DISPOSAL SYSTEM

	Reservoir #1															Mossy Lake									
Station	Weir Surface					Weir 10 Ft.					TP3					Gate					East Arm				
Year	57	58	59	60	61	57	58	59	60	61	57	58	59	60	61	57	58	59	60	61	57	58	59	60	61
Zooplankton																									
Protozoa																									
<u>Paramecium</u>	X	X	X	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Other Ciliates				X	X									X	X	X			X		X	X		X	
<u>Vorticella</u>																						X			
<u>Stentor</u>																		X							
<u>Heliosphaerum</u>																	X			X		X		X	
Rotifera	X	X			X	X						X	X	X	X	X	X	X	X	X	X	X	X	X	X
Gastrotricha																		X	X		X				
Arthropoda																									
Hydracarina													X									X	X		
Crustacea																									
Ostracoda																			X						
Cladocera																									
<u>Daphnia</u>																			X						
Copepoda																									
<u>Cyclops</u>																						X	X		
Other Copepods		X															X	X							
Insecta																									
Bristle Worm (Diptera Larva)																		X							

B. Effect of Rainfall on the *Chlorella* Population

Rainfall was measured at the plant for the period of the study (Fig. 5). There was a heavy monthly rainfall with only three months when rainfall was less than an inch. The highest rainfall in a month was 17.5 inches and the average monthly rainfall was 5.8 inches. There were several floods in spring and summer periods. Peaks were reached in each season of the year at about the same time each year. A winter peak occurred in January or February. The spring was in March or April. There was a summer peak in June or July. The fall peak appeared in November. Runoff after a heavy rain on the watershed significantly increased the quantity of effluent released from the first impoundment.

Rainfall was thought to have a possible effect on the *Chlorella* population since a great change in the lake volume always occurred after rainfalls of more than an inch. At such times the gates might have had to have been opened completely in order to maintain the required level.

Two-way tables were constructed in order to make orthogonal comparisons of the effect of rainfall on *Chlorella* populations during periods without rainfall against periods with rainfall (Tables III and IV). According to Steel and Torrie (1960), orthogonal comparisons permit independent analysis of samples. Factors which have the same effect on each sample are nullified, and analysis is made of factors which have different effects on the samples. This is a useful procedure where interaction of a number of environmental factors occurs. Test periods for comparison were selected from the same season and with water released in the same manner, either over the spillway or below

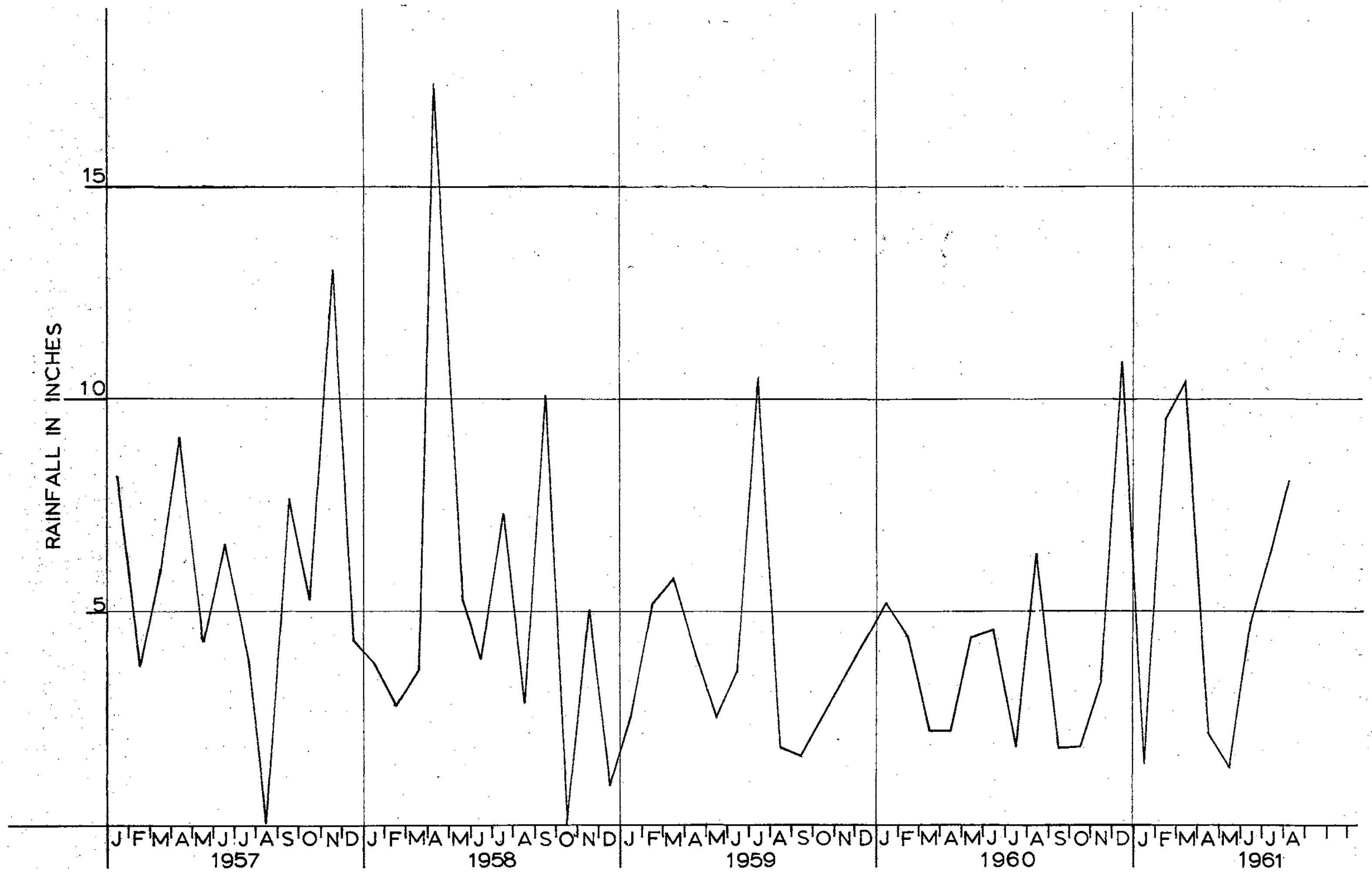

Fig. 5 Rainfall by month on water shed.

the surface. Periods selected for the effect of rainfall had at least an inch of rainfall in the four days preceding the test period and at least an inch of rain in the four days prior to each sampling date. Periods selected for the effect of no rainfall had no more than one inch of rainfall in the four days prior to the test period and no more than one inch of rainfall during the test period.

In Table III, winter, 1961, with no rainfall, is compared with winter, 1961, with rain. Winter Chlorella populations were significantly larger in number when there was no rain than when there was rain.

TABLE III

CHLORELLA POPULATION IN FEBRUARY, 1961

Water released over spillway			
Rain		No Rain	
Date	*	Date	*
2/22	15	2/3	71
2/24	8	2/6	106
2/27	19	2/8	76
3/1	24	2/15	56
Mean	16.5		77.25

* Chlorella in 1/16 cu. mm.

Analysis of Variance

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	7	8836.13		
Treatment	1	7381.12	7381.12	30.42
Error	6	1455.75	342.63	

Tab F 1,6 at 5% level 5.99

Chlorella populations were compared between summer, 1958, when there was rain, and summer, 1961, when there was no rain (Table IV). Summer Chlorella populations were significantly larger when there was no rain than when there was rain.

TABLE IV

CHLORELLA POPULATIONS IN JUNE, JULY, 1958, 1961

Water released below the surface			
1958 (rain) Date	*	1961 (no rain) Date	*
6/16	61	6/27	114
6/18	25	6/29	118
6/20	70	7/3	109
6/23	60	7/5	113
Mean	54.0		113.5

* Chlorella in 1/16 cu. mm.

Analysis of Variance

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	7	8303.5		
Treatment	1	7080.5	7080.5	34.74
Error	6	1223.0	203.83	

Tab F 1,6 at 5% level 5.99

Rain affected the Chlorella populations by dilution. Thus, the numbers of Chlorella found in a given volume of water were significantly less after a rain than in a sample taken in the same season when there was no rain. Samples taken to analyze for a treatment effect other than rainfall were seriously biased unless this factor was taken into consideration. Chlorella has a rapid reproduction rate (Allen, 1955) and samples may be taken within a week after a rainfall.

C. Effect of Settling Basins on Chlorella Populations

Data were selected to study the effect of the settling basins on Chlorella populations (Tables V, VI, and VII). Each of the tables was

TABLE V

CHLORELLA POPULATIONS IN THE SUMMER, 1958, 1959

Before Installation of Basin 1958			After Installation of Basin 1961			
	Date	*	Date	*	Date	*
	6/4	118	6/1	112	6/24	77
	6/6	102	6/3	76	6/27	114
	6/9	36	6/5	76	6/29	118
	6/11	72	6/7	76	7/13	109
Mean		82		85		106.5

* Chlorella in 1/16 cu. mm.

Analysis of Variance

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	11	7.127		
Treatment	2	1,298.5	649.25	1.002
Error	9	5,828.5	647.11	

Tab F 1,9 at 5% level 4.26

composed of population data from three periods in a season. Intervals for study were selected with no more than one inch of rainfall in the four days prior to the test interval and in which no more than one inch of rain fell during the test. These data also permitted comparisons between years and within seasons.

Since annual and in-season variations were not significant, analysis of effects of the settling basin could be made.

In Table V a comparison was made of the summer Chlorella populations in June, 1958, before the settling basin was placed in operation and two Chlorella populations in June, 1961, after the settling basin was in operation. There were no significant differences between these populations, which indicates that operation of the settling basin had no effect on summer algal populations.

In Table VI comparisons were made of winter Chlorella populations in 1958, before the settling basin was installed, and 1959-60, after the settling basin was installed. There was no real difference in winter Chlorella population due to the settling basin.

In Table VII a comparison was made of Chlorella populations in fall, 1957, before the settling basin was placed in operation with a fall period of 1959 after the settling basin was placed in operation. There were no significant differences in the fall Chlorella populations of 1957.

Chlorella populations were significantly less in 1959 than in 1957, due to the effect of variation in plant operation (Table XIII) rather than the operation of the settling basin.

TABLE VI

CHLORELLA POPULATIONS, WINTER, 1958, AND 1959-1960

No Settling Basin		Settling Basin			
1958		1959		1960	
Date	*	Date	*	Date	*
12/12	52	12/2	18	1/11	32
12/15	21	12/4	24	1/13	14
12/17	27	12/7	11	1/15	10
12/19	23	12/9	16		
Mean	30.5		17.25		18.66

*Chlorella in 1/16 cu. mm.

Analysis of Variance

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	10	1408.73		
Treatments	3	426.83	142.28	1.01
Error	7	981.9	140.27	

Tab F 1,9 at 5% level 4.26

The tests show no difference in the Chlorella populations due to the settling basin. The basin has a value in the removal of the sludge before it enters the holding ponds which increases the life of the holding ponds. The removal of this material also improves the appearance of the waste which is one of the purposes of waste treatment. Prior to the installation of the settling basin the bottom of R-1 was covered with a thick ooze, but within two years after this installation the ooze had disappeared. Floating mats of pulp were also eliminated by the introduction of the settling basin. Foaming of soaps remaining in the wastes was decreased with the operation of the settling basin.

TABLE VII

<u>CHLORELLA</u> POPULATIONS OCTOBER, 1957 AND 1959

Before Installation of Settling Basin 1957				After Installation of Basin 1959	
Date	*	Date	*	Date	*
10/4	67	10/28	70	10/19	63
10/7	53	10/30	93	10/21	44
10/9	72	11/1	117	10/23	41
10/11	85	11/4	128		--
Mean	69.25		102		49.3

* <u>Chlorella</u> in 1/16 cu. mm.

Analysis of Variance

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	10	7,834.18		
Treatment	2	5,018.76	2,509.38	7.13
59 vs 57,57	1	2,292.74	2,292.74	6.51
57 vs 57	1	780.05	780.05	2.21
Error	8	2,815.42	351.93	

Tab F 1,9 at 5% level 4.26

Plate I Settling Basin: Upper in operation, lower drained in preparation for dredging.

D. Effect of Variation in the Release of Effluent on *Chlorella* Populations

Tests were designed to compare the effect of releasing water at the surface with release at a sub-surface level. Waste water was released from the first impoundment either at the surface over a spillway, or through a gate 10 feet below the water surface. Two-way tables were constructed for analysis of variance on the effect of the method of release on the *Chlorella* populations (Tables VIII, IX, X, and XI). Test periods were selected in which no more than one inch of rain fell in the four days preceding the period and in which no more than one inch fell in the course of the test. Comparison test periods were restricted to one season.

Two sampling intervals were chosen in the fall of both 1959 and 1960 (Table VIII). Selection of two intervals in each year permitted the comparison of periods within a season. The *Chlorella* population did not change during the season of either year. The *Chlorella* population of 1959 was significantly less than in 1960. Water released at the lower depth resulted in the population being significantly less than with a surface release of water.

In Table IX a comparison was made of *Chlorella* populations in October, 1958, when water was released 10 feet below the surface, with populations in October, 1959 and 1960, when water was released at the surface. The populations of *Chlorella* in the fall were significantly less when the water was released below the surface than when water was released at the surface. The *Chlorella* population was significantly less in 1959 than in 1960 (see also Table XIII).

TABLE VIII

EFFECT OF VARIATION IN WATER RELEASE ON CHLORELLA POPULATIONS, OCTOBER AND NOVEMBER, 1959, 1960

Water Released at 10 Ft. Depth 1959				Water Released over the Spillway 1960			
Date	*	Date	*	Date	*	Date	*
10/19	63	11/18	39	10/19	77	11/4	57
10/21	44	11/20	47	10/21	56	11/6	65
10/23	41	11/23	35	10/24	65	11/9	70
10/26	41	11/25	30	10/26	65	11/11	68
Mean	47.25		39.75		65.75		65

* Number Chlorella in 1/16 cu. mm.

Analysis of Variance

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	15	3,086.94		
Treatments	3	2,894.69	964.90	60.23
59 vs 60	1	478.52	478.92	29.87
59 vs 59	1	28.12	28.12	1.25
60 vs 60	1	00.28	00.28	0.02
Error	12	192.25	16.02	

Tab F 1,12 at 5% level 4.75

TABLE IX

EFFECT OF VARIATIONS IN WATER RELEASE ON CHLORELLA POPULATIONS, OCTOBER, 1958, 1959, AND 1960

Water Released at 10' Depth		Water Released Over Spillway			
1958		1959		1960	
Date	*	Date	*	Date	*
10/20	33	10/19	63	10/19	77
10/22	32	10/21	44	10/21	56
10/24	38	10/23	41	10/24	65
10/27	27	10/26	41	10/26	65
Mean	32.5		47.25		65.75

* Chlorella in 1/16 cu. mm.

Analysis of Variance

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	11	2,841.0		
Treatment	2	2,220.5	1,100.25	15.96
58 vs 59,60	1	840.50	840.50	12.94
58 vs 60	1	2,221.13	2,221.13	32.22
Error	9	620.50	68.94	

Tab F 1,9 at 5% level 4.26

In Table X populations of Chlorella are compared between June, 1961, with the water released at a sub-surface level and in July, 1961, with the water released at the surface. Summer Chlorella populations were larger in water released below the surface than in water released at the surface.

TABLE X

EFFECT OF VARIATIONS IN WATER RELEASE ON CHLORELLA POPULATIONS, SUMMER, 1961

Water Release Over Spillway Date	*	Water Release at 10' Depth Date	*
7/21	33	6/27	114
7/24	83	6/29	118
7/28	56	7/3	109
7/31	77	7/5	113
Mean	62.25		113.5

* Chlorella in 1/16 cu. mm.

Analysis of Variance

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	7	6,836.88		
Treatment	1	5,253.0	5,253.0	19.89
Error	6	1,583.88	263.98	

Tab F 1,9 at 5% level 4.26

In Table XI comparisons were made between two late summer samples in 1960 when water was released at 10 foot depth and the summer of 1961 with water release over the spillway. A period within period variance was analyzed for 1960. More Chlorella were present during release at the sub-surface level than when the water was released at the surface. Comparison of the two periods in 1960 showed no difference in populations.

TABLE XI

EFFECT OF VARIATIONS IN WATER RELEASE ON CHLORELLA POPULATIONS
AUGUST, SEPTEMBER, 1960, 1961

Water Release Over Spillway 1960				Water Release at 10' Depth 1961	
Date	*	Date	*	Date	*
8/26	59	9/14	27	8/14	75
8/29	31	9/16	35	8/16	76
8/31	43	9/19	75	8/18	76
9/2	45	9/21	56	8/21	76
Mean	44.5		40.75		75.75

* Chlorella in 1/16 cu. mm.

Analysis of Variance

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	11	4,422.67		
Treatment	2	2,504.17	1,252.09	6.56
61 vs 60,60	1	2,501.04	2,501.04	13.14
60 vs 60	1	312.55	312.55	0.02
Error	9	1,718.5	190.94	

Tab F 1,9 at 5% level 4.26

All tests showed significant differences in Chlorella populations resulting from variations in water release. Raising the water level to permit discharge over the spillway resulted in higher Chlorella populations in the fall but reduced populations in the summer.

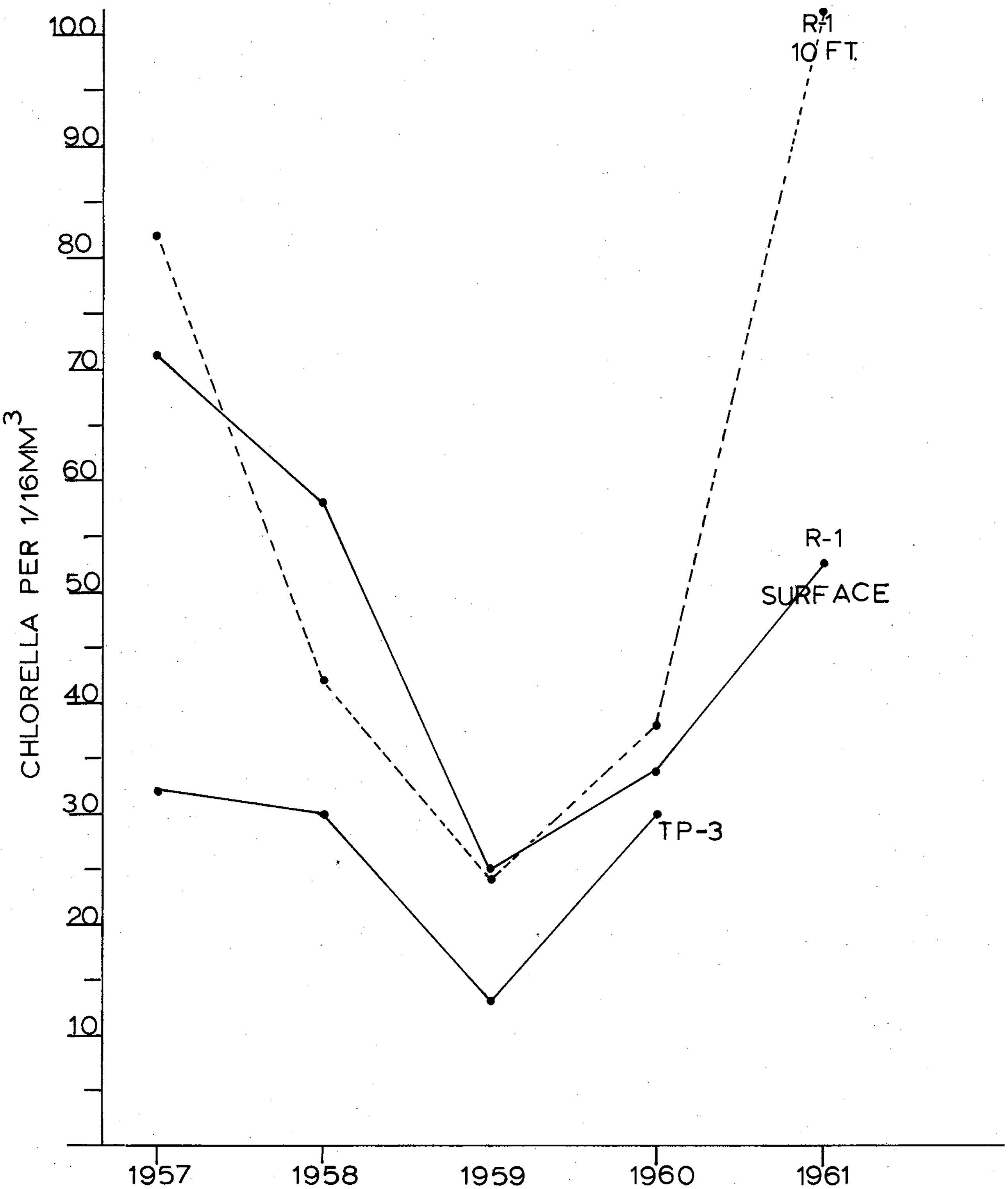

Fig. 6 Numerical Succession in Chlorella in System and Time

E. —In-season, Seasonal, and Annual Variations in Chlorella Populations

Annual differences in Chlorella populations before the settling basin was placed in operation were compared (Table XII). Two intervals in September, 1957, were compared with an interval in September, 1958. Water was released through the gate at the base of the dam.

Chlorella populations in 1957 and 1958 were not significantly different. The Chlorella populations of two summer periods were not significantly different.

TABLE XII

CHLORELLA POPULATIONS IN SEPTEMBER, 1957 AND 1958

1957		1957		1958	
Date	*	Date	*	Date	*
9/9	22	9/16	20	9/22	15
9/13	55	9/19	17	9/25	47
9/14	20	9/21	15	9/27	32
9/15	17	9/22	21	9/29	38
Mean	23.5		18.25		33

* Chlorella in 1/16 cu. mm.

Analysis of Variance

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	11	1,974.92		
Treatment	2	457.17	228.58	1.35
Error	9	1,517.75	168.63	

Tab F 1,9 at 5% level 4.26

All data on Chlorella populations from the weir station of the upper holding pond were grouped into seasonal and yearly periods (Table XIII). Missing data were computed by the missing plot technique (Steele & Terrey, 1960).

There was no significant difference in the Chlorella population between 1957, 1958, 1960, and 1961. Chlorella populations of 1959 were found to be significantly smaller than in the other years. Changes in plant operation may have caused this difference. These changes included installation of a new evaporator in the tall-oil plant and work stoppages in the mills. The organic load (BOD-COD) was less in 1959 than for all other years of the study (Fig. 7). The additional evaporator produced a higher quality of effluent because there was a higher percentage of recovery of re-useable products. When the work stoppages occurred the volume of effluent released remained the same but the BOD-COD load was decreased. Rainfall was not considered a factor since in 1959 there was less precipitation than in the other years of the study (Fig.5). The BOD-COD load of the total plant effluent was higher in 1960, an indication that the new evaporator had less effect than did the work stoppages (Fig. 7).

In-season variations of Chlorella populations were not significant (Tables V, VI, VII, VIII, and XI). Annual variations in Chlorella populations except in 1959 were not significant (Tables V, VI, and VIII). Significant differences between the Chlorella population of 1959 and that of other years were expressed (Tables VII, IX, and XIII). Chlorella populations did not vary significantly between seasons.

Chlorella populations did not vary significantly from season to season. Seasonal effects result from a combination of available light and temperature. Light penetration into the water of the holding pond

TABLE XIII

AVERAGES OF ALL CHLORELLA POPULATIONS AT THE OUTLET OF UPPER RESERVOIR

Year	Spring	Summer	Fall	Winter	$\bar{x}$
57	55.71*	41.44	119.45	76.13	79.00
58	63.19	57.73	30.38	31.88	45.80
59	18.75	19.11	28.26	29.3	23.86
60	15.33	41.13	55.92	44.10	39.12
61	31.48	66.89	48.42*	48.75*	38.34
Mean	32.41	45.26	58.5	45.35	45.38
Settling Basin					
Absent	63.19	49.59	74.91	54.0	
Present	21.85	42.37	42.0	36.7	

Analysis of Variance

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	19	11,079.94		
Years	4	5,139.40	1,284.85	3.10
59 vs rest	1	2,489.24	2,489.24	6.00
57,58 vs 60,61	1	959.30	959.30	2.31
57 vs 58	1	1,500.15	1,500.15	3.62
60 vs 61	1	190.71	190.71	0.46
Seasons	3	966.74	322.25	0.78
Error	12	4,973.60	414.48	

Tab F 1,12 at 5% level 4.75

Tab F 4,12 at 5% level 3.26

* Missing data

was four inches because of color and turbidity so that the amount of available light was reduced at all times. *Chlorella* is less sensitive to light intensity (Allen, 1955) and thus is more able to survive under extreme conditions than most other algae.

Water entered the reservoir at a temperature of 35°C. at all seasons of the year. Seasonal fluctuation of water temperature was reduced at all points in the reservoir and was virtually eliminated in the upper end of the reservoir. Uniform waters might be expected to promote populations of organisms throughout the year.

F. Open Water *Chlorella* Collections

Studies of the *Chlorella* populations of the open water at R-1 were made to determine whether there were variations at different places in the reservoir. Analysis was made of the six stations (Fig. 2) for all collections after installation of the settling basins (Table XIV). No significant difference was found in the *Chlorella* populations due to the location of the collecting stations. Variation in populations of *Chlorella* between stations was not significant as the variation between points was less than variation due to the sampling period. Collections made at the outlet could thus be used to find differences in populations due to change in treatment.

TABLE XIV

COMPARISON OF *CHLORELLA* POPULATIONS AMONG OPEN WATER STATIONS

Source	Degree of Freedom	Sum of Squares	Mean Square	F
Total	119	401,080.75		
Stations	5	11,768.77	2,353.75	26.89
Treatments	9	141,540.21	15,726.69	26.89
59,61 vs 59,60,61	1	28,305.41	28,305.41	48.0
59 vs rest	1	2,706.82	2,706.82	4.5
60,60,60 vs 59	1	2,562.67	2,562.67	4.05
59 vs 61	1	12,535.51	12,535.51	21.15
60 vs 61	1	13,378.78	13,378.78	22.5
Error SxP	45	26,321.57	548.92	

Tab F 1,45 at 5% level 4.06

G. -Chemical Components of the Effluent

Concentration of chemical constituents in the effluent generally decreased as the waste progressed through the system. Biochemical oxygen demand and chemical oxygen demand were examples of factors which decreased with the progressive improvement of the effluent. COD-BOD are shown in Figure 7 . This figure is based on more than 5000 determinations during the five year period. At the point of exit from the plant, TP-1, (Fig. 1) oxygen consuming capacity of the waste was high but after the waste was exposed to natural purification processes in the two holding ponds the oxygen demand properties of the waste were largely satisfied.

There was little change in pH through the system. The waste at the plant exit had an average pH of 7.8 with a range of 6.6 to 8.6. The effluent at the lower end of the system had an average pH of 7.8 with a range of 7.2 to 8.2. The receiving stream had a lower pH range of 6.9 to 7.5 and was of such volume that dilution to 6.9 was immediate after confluence. This pH range was well within the limits acceptable for aquatic life (McKee et al, 1957).

Total soda concentration was safely under the amount considered detrimental to aquatic life (McKee, 1957). The average sodium concentration was 191 ppm and was not changed as the waste passed through the holding ponds. The average total sodium concentration for TP-1 was 191, for R-1 was 191, and for TP-3 was 183. All concentrations were of the same magnitude.

Total dissolved solids in the waste water were reduced in passage downstream. Total dissolved solids averaged 1,330 ppm at the point of exit from the plant grounds. The settling basin installed in 1959 made a significant difference in the removal of dissolved solids as well as in sludge removal since the average total solids of the wastes in R-1 reservoir was 940 ppm. There was little change in the total solids in Mossy Lake at TP-3, point of exit from the system, where the average total solids was 867 ppm.

Water color is a quality of esthetic importance and functional value in many industrial operations. In papermills, for example, color lowers the grade of the product. Papermills wastes are noted for color. Color comparisons were made to determine the extent of color change induced by passage through the series of holding ponds. The waste at the exit from the plant grounds was not measured, since dyes

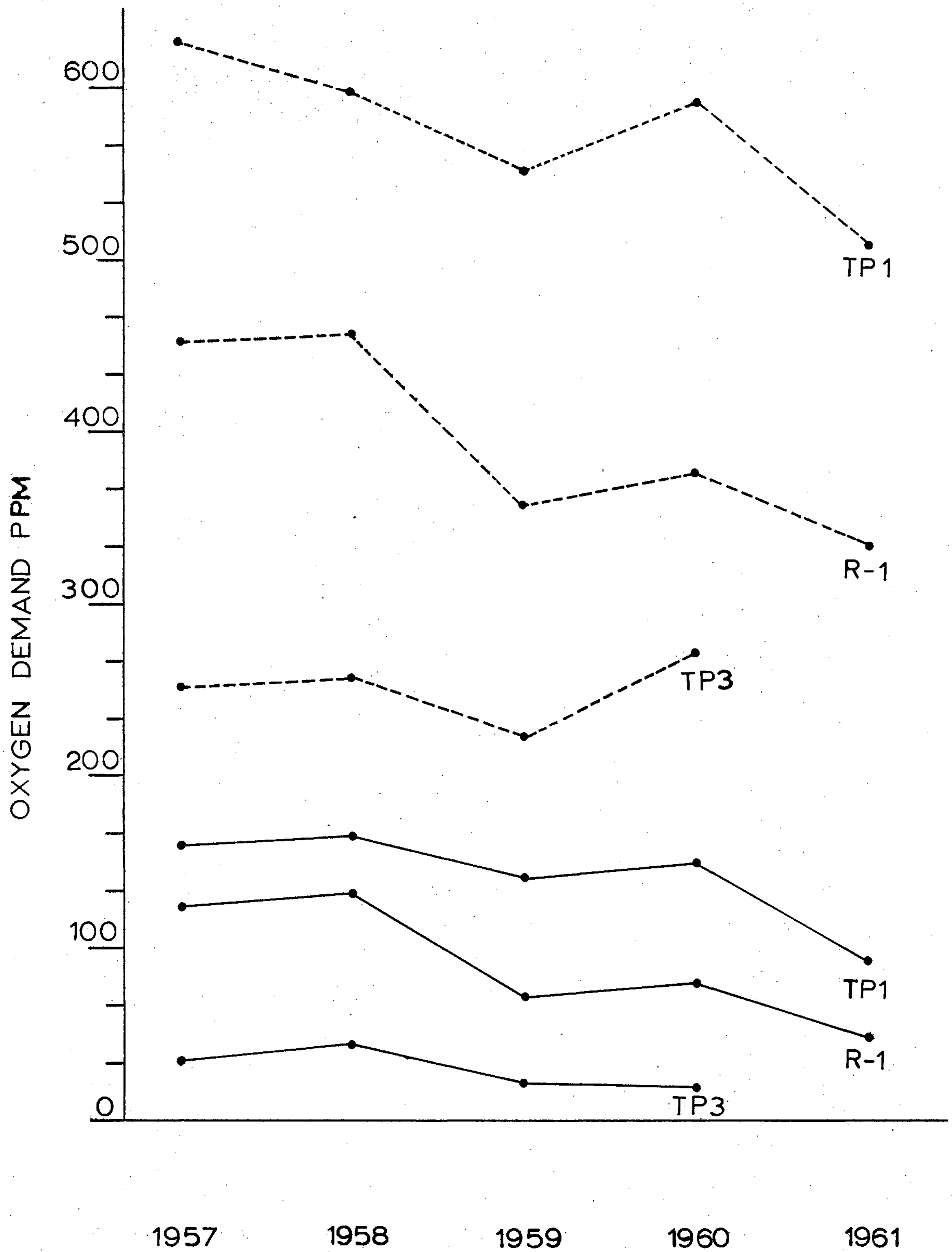

Fig. 7 BOD-COD Changes in Oxygen Consumption: broken lines represent COD, solid lines represent BOD

used in paper processes produced variable effects. The average color index was 525 ppm in R-1. In Mossy Lake the color index dropped to an average of 290 ppm. This showed that color was lost from the waste in holding ponds.

Hydrogen sulfide has long been a troublesome product in holding ponds in crowded areas. Crude measurements showed that introduction of the settling basin had significant effect in the removal of this compound. Prior to installation of the settling basin the gas was easily detected both at the upper and lower holding ponds. Afterwards only a faint indication was recorded on the lead acetate paper.

IV. SUMMARY

A study was made on the algal populations of the waste impoundment system of a Kraft papermill. Population variations in the reservoir were insignificant, so that it was considered feasible to base analysis on samples taken at the outfall.

The Chlorella populations in the effluent of papermills indicated the recovery stage of the effluent. Comparison of Chlorella population data was restricted so that only one variable was examined at a time.

Chlorella was the dominant alga at all points in the system, but its numbers were reduced in the lower section.

Rainfall was an important factor governing population density. Chlorella populations were less dense after rains as a result of dilution.

Water was released from an upstream reservoir at the surface or through a culvert 10 feet below the surface of the water. Release of the water over the spillway produced a significant difference in the Chlorella population. Chlorella numbers were increased in the summer when the water was released at the surface of R-1, but the number of Chlorella were decreased by surface release in the fall. Raising the water level to permit waterflow over the spillway produced a change in the holding time as there was more water volume in the lake. An increase of holding time in the system would probably improve the quality of water released into the receiving stream, since two sampling points

at the second holding pond where the water was by-passed and had a longer retention period contained more genera of algae than were found at the end of the system. Changes in the number of genera of algae as the effluent progressed downstream indicated changes in environmental conditions.

No difference in Chlorella populations was found between years with the exception of 1959 when plant operations produced an unusually light BOD-COD load with a correspondingly lower Chlorella population.

Sampling periods within the same season and with similar treatment had no significant differences in Chlorella populations.

The installation of a settling basin for sludge removal resulted in no significant change in the number of Chlorella.

Holding time amounted to about 45 days in actual operation.

Water entered the upper holding pond at about 35°C. at all seasons. Light penetration of the water of the upstream reservoir was about four inches, and in the lower reservoir the light penetration was about eight inches. The pH of the effluent averaged 7.6 with a range from 7.2 to 8.2 in Mossy Lake and a range at the plant of 6.6 to 8.6. The COD-BOD load was reduced as the effluent moved downstream indicating a reduction in organic components.

SELECTED BIBLIOGRAPHY

Allen, M. B., 1955. General features of algal growth in sewage oxidation ponds. State Water Pollution Control Board. Sacramento, Calif. Publ. No. 13, 48 pp.

A.P.H.A. (American Public Health Association), 1960. Standard methods for examination of water and wastewater. 11th Ed. 626 pp.

Gaufin, A. R., and C. M. Tarzwell, 1956. Aquatic macro-invertebrate communities as indicators of organic pollution in Lytle Creek. Sewage and Industrial Wastes, Vol. 28, No. 7.

McKee, J. R., and Advisory Committee, 1957. Water quality criteria (including addendum no. 1) State Water Pollution Control Board. Sacramento, Calif. Publ. No. 3, 2nd Ed. 672 pp.

Minter, K. W., unpublished. Population dynamics of plankton in oil refinery effluent holding ponds.

Oswald, W. J., H. B. Gotaas, C. G. Golueke, and W. R. Kellen, 1957. Algae in waste treatment. Sew. Ind. Wastes. 29:437-455.

Pennak, R. W., 1953. Fresh-water invertebrates of the United States. Ronald Press Co., New York. 740 pp.

Prescott, G. W., 1954. How to know the fresh-water algae. Wm. C. Brown Co., Dubuque, Iowa. 198 pp.

Silva, P. C. and G. F. Papenfuss, 1953. A systematic study of the algae of sewage oxidation ponds. California State Pollution Control Board. Sacramento, Calif. Publ. No. 7.

Smith, G. M., 1950. The fresh-water algae of the United States. 2nd Ed. McGraw-Hill Co., New York. 719 pp.

Steele, R. G. D., and J. H. Torrie, 1960. Principles and procedures of statistics. McGraw-Hill Co., New York. 472 pp.

Van Horn, W., 1949. A study of kraft pulping wastes in relation to the aquatic environment. In Moulton and Hitzel. 1949. Limnological aspects of water supply and waste disposal. Amer. Assoc. Adv. Sci., Washington, D.C. pp. 49-55.

Welch, P. S., 1949. Limnological methods. The Blakiston Co., Philadelphia, Pa. 370 pp.

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