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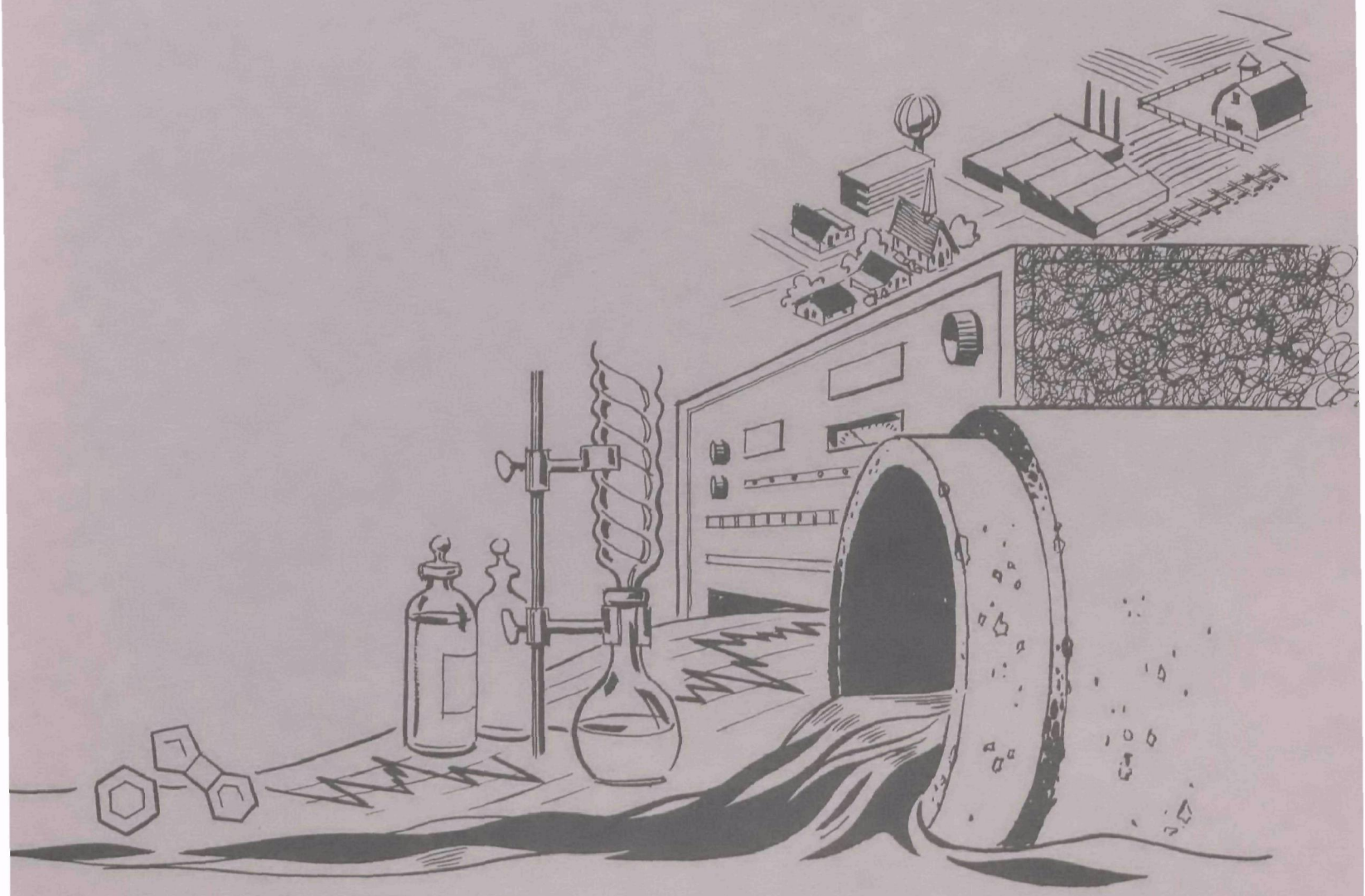

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A MICROBIOLOGICAL SURVEY IN LAKE ERIE NEAR CLEVELAND, OHIO

by

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for the

Office of Research and Monitoring

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ABSTRACT

For several years the Crown Water Treatment Plant in Cleveland, Ohio has experienced periodic taste and odor problems and the present investigation was concerned with the role that microorganisms play in this problem. During June, July and August of 1971 collections of fungi, bacteria and algae were made near the intake of the Crown Treatment Plant.

The studies showed that fungi and bacteria played little, if any, role in the taste and odor problem at the Crown Plant. However, a number of algae which have been reported to induce taste and odor in water supplies were identified in the present study. Those taste and odor algae which were found in relative abundance included: *Ceratium* sp., *Coelosphaerium* sp., *Dinobryon* sp., *Fragilaria* sp., *Pediastrum* sp., *Staurastrum* sp., *Tabillaria* sp., and *Mougeotia* sp.

There was no evidence that benthic organisms played any significant role in the taste and odor problem experienced at the Crown Treatment Plant.

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SECTION I

CONCLUSIONS

1. Satisfactory sampling procedures for both phytoplankton and benthic organisms were developed.

2. It was concluded that bacteria and fungi play little, if any, role in the taste and odor problem at the Crown Treatment Plant.

3. No evidence could be found that benthic organisms play a significant role in the development of tastes and odors at the Crown Treatment Plant.

4. Certain phytoplankton collected did produce earthy odors.

SECTION II

RECOMMENDATIONS

The problem of taste and odor in Lake Erie and, more particularly, at the Crown Treatment Plant is undoubtedly complex in origin, and if an understanding of the problem is to be achieved, a more comprehensive approach than the one utilized in the summer of 1971 is needed.

This particular problem needs more adequate funding if a thorough job is to be done. Collections should be made more frequently and over a longer period of time than was possible in the present study.

If the problem were continued, a shift in the kinds of personnel employed would be desirable. The present study has demonstrated that bacteria and fungi do not play a significant role, therefore, continued expertise in these areas is not necessary. A person skilled in phytoplankton work, including skills in identification and culturing, will still be needed. It is recommended that an organic chemist broadly trained in the isolation of trace amounts of organic constituents could be profitably employed. A sanitary engineer could be of some help to the team, particularly if he has knowledge of the area.

It is recommended that at least some of the personnel have sufficient diving skills to enable them to work in depths of water up to sixty feet.

It is recommended that a mobile laboratory be placed at the site so that analyses could be continuous and that time is not lost in transporting samples to adequate laboratories. The mobile laboratories should be under the control of the EPA and should be released for stipulated periods and then returned to a central pool for reassignment.

If it were at all possible, the EPA should also have a facility of this sort for boats. One of the amazing things in the recent study was the fact that boats of the size needed for this work were extremely difficult to locate.

SECTION III

INTRODUCTION

The western suburbs of the city of Cleveland, Ohio have had problems with taste and odor in the municipal water supply for the past several years. This condition was particularly severe during the summer of 1966 and again during the summer of 1967.

The western suburbs of Cleveland are served by the Crown Water Treatment Plant, which is a rapid sand filter plant with a design filtering capacity of 50 mgd. The raw water intake for the Crown Plant is located 2.5 miles offshore, 46 feet below the surface of Lake Erie. The circular crib is 10 feet high and has a diameter of 60 feet. Initially, the intake was located four feet above the bottom and at the center of the crib.

Potos (1) and Kleveno, Braideck and Gehring (2) concluded that most of the taste and odor complaints occurred when the Crown Plant raw water intake was located in the hypolimnion. These same workers showed that the appearance of a hypolimnion in the vicinity of the Crown Plant was dependent upon prevailing winds from a southerly direction. When the wind direction is southerly, the surface waters of Lake Erie are pushed to the northern shores with the result that the hypolimnion rises in the south and becomes depressed in the northern area.

It was assumed that the problem at the Crown Plant could be solved by moving the intake so that it would always be above the hypolimnion. However, when the intake was placed above the hypolimnion, it was found that the taste and odor problems continued. This suggested that there was a break in the intake line and investigation revealed that this was indeed the case. During the course of this investigation, plans to repair the break in the intake line were being carried out and it is assumed that this will solve to a large extent the taste and odor problem existing at the Crown Plant. This optimism seems warranted because other nearby plants which draw their water only from the epilimnion have not experienced the problems found at the Crown Plant.

The taste and odor problems at the Crown Plant during the summer of 1971 were not particularly severe and this could be attributed to environmental conditions which precluded the formation of a persistent hypolimnion.

In the study reported by Potos (1), sampling was done for the most part in the vicinity of the Baldwin Water Treatment Plant intake. The Baldwin Crib is located 7 miles east of the Crown Plant Crib. The Baldwin site has definite advantages because the Crib is raised and sampling is more reproducible. In the present study, however, it was felt that the collections should be made as close to the Crown Crib as possible and, therefore, all collections were made in the immediate vicinity of the Crown Crib as well as within the Crib. Some collections for bacterial analyses were made in the vicinity of a marker buoy approximately two miles offshore.

SECTION IV

MATERIALS AND METHODS

This section will be divided into three parts: the first part dealing with the algae; the second with the fungi; and the third with the bacteria.

Plankton samples were taken at various depths in the vicinity of the Crown Point Intake. Samples initially were taken with a six liter plankton bottle and immediately concentrated by pouring the samples through a No. 20 mesh plankton net. The concentrated material was collected in a vial attached to the net. In later experiments the water samples from each depth were increased to 12 liters.

Benthic algae samples were collected by scuba divers who pushed a 10" x 7" open plexiglass box into the soft muck bottom. A bottom plexiglass plate was slipped underneath the box and the isolated muck samples brought to the surface.

Periphyton samples were collected from the cement walls and rocks at the base of the crib. All samples were fixed in a preservative, stored in an ice chest, and transported to the laboratory in Storrs, Connecticut, where final analyses were made.

Water samples, taken at the same depths as the plankton samples, were collected with six liter plankton bottles for chemical analyses. The pH, alkalinity, carbon dioxide, and oxygen concentrations were analyzed immediately aboard the vessel. The remaining water from each depth was stored in glass bottles, placed in an ice chest, and the remaining analyses carried out in Storrs, Connecticut.

All of the above experiments concerned with chemical analyses were performed with the Hach Kit.

Plankton counts, in the initial experiments, were determined by a millipore filter method. However, this method proved to be unsatisfactory. All later samples were analyzed by the Sedgwick Rafter Cell Method. The plankton were identified and counted using a 12.5x eyepiece and a 10x objective. Filamentous algae were counted by cell counts; colonial and all other species were counted as individuals. Plankton data are reported as organisms per liter.

The fungi were collected by taking an aliquot of water from the six liter plankton bottle at each of the depths sampled. The collected material was placed in sterile Blake bottles and immediately placed on ice in an ice chest. The samples were then analyzed at the Connecticut laboratories.

Aliquots (2 ml), either diluted or undiluted, were placed on various media (see Table 1) and then three replicate flasks for each depth were

incubated at 20°C and 25°C for varying periods of time. As soon as cultures had grown out sufficiently, identifications were made.

The only exception to the above procedure was in those cases where hemp seed and hair were used to trap aquatic molds. In those cases one or two sterile hemp seeds and a few strands of human hair were placed in a sterile petri dish. Water collected from one of the sampling depths was then poured into the petri dishes (approximately 15 ml of lake water added) and the petri dishes incubated in the usual fashion. The procedure described above was used for water collected at each depth sampled.

Table 1. Formulation of media used in the isolation and culture of fungi.

Rose Bengal Agar

Ingredients	Amounts
Neopeptone	5.0 g
Dextrose	10.0 g
Rose Bengal	0.035 g/l
Aureomycin HCl	35.0 g/l
Agar	20.0 g
Water	1000.0 ml

Sabouraud Dextrose Agar

Ingredients	Amounts
Neopeptone	10.0 g
Dextrose	40.0 g
Agar	10.0 g
Water	1000.0 ml

Littman-Oxgall Agar

Ingredients	Amounts
Neopeptone	10.0 g
Dextrose	10.0 g
Oxgall	15.0 g
Agar	20.0 g
Crystal-violet	0.01 g
Water	1000.0 ml

Malt Extract Agar

Ingredients	Amounts
Yeast extract	3.0 g
Malt extract	3.0 g
Neopeptone	5.0 g
Dextrose	10.0 g
Agar	20.0 g
Water	1000.0 ml

Lindegren Yeast Agar

Ingredients	Amounts
Yeast extract	5.0 g
Proteose peptone No. 3	3.5 g
Dextrose	40.0 g
Potassium acid phosphate	2.0 g
Magnesium sulfate	1.0 g
Agar	20.0 g
Water	1000.0 ml

The samples used in the bacterial analyses were collected at the crib and at a site approximately one-half mile south of the crib. For the analysis of aerobic bacteria, a Nisken Sampler was used (3). For the analysis of anaerobic bacteria, collections were made by putting approximately 25 ml of water obtained from the plankton bottle into sterile one-ounce prescription bottles containing one ml of Bacto-fluid thioglycollate medium. These bottles were placed in a desiccator which was evacuated as soon as the ship reached the shore. In both of the situations described above collections were made at the surface and at depths of 20 and 40 feet.

The Niskin bags, along with the desiccator, were stored in an ice chest until examined at Storrs. Sediment samples were collected in the plexiglass sampler previously described and the samples for bacteriological examination were removed with a sterile spatula and placed in sterile bottles. These bottles were placed in the desiccator containing the anaerobic water samples. Spread plates (4) on Bacto-Plate Count Agar (5) were incubated at 35°C, 20°C and 5°C for 24 hours, 48 hours and 4 weeks respectively. Appropriate dilutions were made using sterile distilled water. Total and fecal coliform determinations were made on membrane filters (HAWG, 0.45 u, Millipore) according to "Standard Methods for the Examination of Water and Waste Water" (6).

The anaerobic samples were plated on plate count agar and the plates were incubated in an anaerobic jar evacuated with a BBL Gas-Pak at 20°C. Samples were also placed in anaerobic sulfate broth consisting of yeast extract, 1 g; neopeptone, 1 g; sodium lactate, 8.0 g; sodium sulfite, 0.1 g; ammonium sulfate, 0.1 g; magnesium sulfate, 0.1 g; ascorbic acid, 0.1 g; ferrous ammonium sulfate, 0.1 g; 2 ml of potassium dibasic phosphate; and 1.0 liter of distilled water. The media was dispensed into sterile test tubes, the water sample added and then the tubes were covered with sterile paraffin and incubated at 20°C for 4 weeks. Water samples from the Niskin bags were aseptically added to flasks containing media for nitrifying and iron bacteria. The media for the nitrifying bacteria consisted of $(NH_4)_2SO_4$, 200 mg; K_2HPO_4, 50 mg; chelated metals solution, 1 ml; $CaCO_3$, 0.3 g; Phenol Red, 1.0 ml of 0.5% aqueous solution; distilled water, 1000 ml ($CoCl_2 \cdot 6H_2O$, 0.004 g; $CuSO_4 \cdot 5H_2O$, 0.0004 g; $FeCl_2 \cdot 6H_2O$; 1.0 g; $ZnSO_4 \cdot 7H_2O$; 0.3 g; $MnSO_4 \cdot H_2O$; 0.6 g; $Na_2MoO_4 \cdot 2H_2O$, 0.15 g; EDTA, 6 g; made up to one liter with glass-distilled water). The liquid medium for iron bacteria (Lieoke) consisted of ammonium sulfate, 1.5 g; potassium chloride, 0.05 g; magnesium sulfate, 0.05 g; potassium monobasic phosphate, 0.05 g; calcium nitrate, 0.01 g; and

distilled water, 1000 ml. These flasks were incubated at 20°C for 4 weeks. At the end of this time period the flasks of Lieoke's medium were streaked out on Waksman's medium containing ammonium sulfate, 0.5 g; magnesium sulfate, 0.5 g; potassium monobasic phosphate, 0.5 g; sodium nitrate, 0.5 g; calcium chloride, 0.2 g; agar, 18.0 g and distilled water, 1000 ml; this media was placed in test tubes and when the plates were poured 1.0 ml of a 15% solution of ferric ammonium citrate was added. Samples were also filtered through membrane filters for incubation on yeast medium consisting of nutrient agar, 2.3 g; glucose, 1.0 g; yeast extract, 0.1 g; malt extract, 0.2 g; chloromycetin, 100 mg; distilled water, 100 ml. These plates were incubated at 20°C for one week.

SECTION V

EXPERIMENTAL RESULTS

As in the Materials and Methods section, the results for each group of organisms will be considered separately and the chemical data will be included with algae result section.

During the three months of sampling (June-August) no vertical pattern of distribution was noted in quantitative analysis of the plankton flora. Wright and Tidd (7) also observed this same phenomenon. Yearly variation in the production of phytoplankton have been observed by Chandler and Weeks (8) in Lake Erie and they attribute this to temperature, rate of eastward flow of water and nutrients emptying into the lake from streams.

In June the maximum concentration of phytoplankton (organism per liter) was considerably lower than that observed in July and August. Maximum concentration occurred at the surface with 13,674 organisms per liter. Similar concentrations were observed at the 3 and 9 meter depths. The lowest concentrations were observed at the 6 and 12 meter depth (Table 2).

Table 2. Vertical sampling of plankton flora and fauna (organisms per liter) taken from Lake Erie at the Crown Point Intake.

6-19-71

Organisms per liter	Surface	3.05M	6.10M	9.15M	12.20M	13.73M
Asterionella formosa	261	614				
Colonial greens	87		174	87	87	
Fragilaria crotonensis	9122	6137	3819	3815	6631	
Mougeotia sp.	3682	5262	2752	8175	998	
Pediastrum duplex		701	87	632	362	
Staurastrum sp.		87				
Tabellaria fenestrata	522			270		
Total organisms per liter	13,674	12,802	6,832	12,979	8,078	
Cladocerans	87			87		
Copepods	435	87	618	453	179	
Rotifers	87	87		87		
Total organisms per liter	609	174	618	609	179	

In July the concentration of phytoplankton (organisms per liter) was considerably higher than that observed in June (Table 3). The highest concentration occurred at the surface with 225,682 organisms per liter. The concentration of phytoplankton decreased progressively with depth to a minimum of 44,522 organisms per liter at the 12 meter depth.

In August, the total concentration of phytoplankton observed at various depths was similar to that seen in July. Maximum concentration occurred at 12 meters with 208,419 organisms per liter (Table 4).

From June to August, two species of phytoplankton appeared to be dominant at all depths sampled. These species were Fragilaria crotonensis and Mougeotia sp. The relative abundance of these two species at all depths sampled ranged from 72% to 96%. In July and August, Ceratium hirundinella and Tabillaria fenestrata appeared to increase in numbers over that observed in June.

The fauna observed during the sampling period of July through August appeared to be relatively low with Copepods being dominant in June and Rotifers dominant in July and August.

Based on monthly sampling, the total number of algal species observed increased from June to August. The number of species observed increased from 7 in June to 26 in August. In June and July, the number of species appeared to be generally uniform with increased depth. In August, the number of species appeared to increase to a depth of 12.20 meters, after which a decline in species numbers was observed (Tables 4-6).

In terms of the total number of species observed at each depth, the results of the July and August analysis indicated that the Chlorophyta (green algae) were the dominant phytoplankton group present, totaling from 45% to 70% of the community. During both sampling periods, the ratio of green algae to total species was less at the surface than that observed at all other depths. Similar results were obtained in the June sampling.

From direct observation and sampling of the bottom mud, no benthic algae were observed during June through August. The benthic mud samples were brought to the laboratory and placed in a culture room in an attempt to promote growth. Samples were left in a lighted culture room (100 f.c.) for a period of a week to ten days during which time no algal growth was observed.

No periphyton algal community of any significance was observed growing on the rocks or walls of the water intake crib, although a few clumps of Cyanophyta (blue-green algae) were observed on the walls during the August sampling.

Prior to the July sampling of the waters around the Crown Point Intake, information was received indicating that odor and taste problems were becoming very obvious in the Cleveland, Ohio area. In comparing the phytoplankton data for June and July, no drastic shift in the community

Table 3. Vertical sampling of plankton flora and fauna (organisms per liter) taken from Lake Erie at the Crown Point Intake.

7-17-71

Organisms per liter	Surface	3.05M	6.10M	9.15M	12.20M	13.73M
Anabaena sp.	1089	442		221	442	
Asterionella formosa	2397		663			
Ceratium hirundinella	7848	4861	3756	1989	884	
Coelastrum microporum			110	221	221	
Cosmarium sp.		110	110			
Dictyosphaerium sp.	870	1789	1105	442	994	
Dinobryon sp.				1989		
Fragilaria crotonensis	43,829	20,107	20,770	10,827	11,490	
Microcystics aeruginosa	216	221				
Mougeotia sp.	162,241	120,864	72,253	45,959	28,945	
Cocystics sp.	1308	1989	1768	663	221	
Pediastrum duplex	1743	442		221	442	
Pediastrum simplex		884	221	110	110	
Scenedesmus sp.			221			
Staurastrum sp.	1525	663	1325	663	663	
Tabellaria fenestrata	2616	221	663	221	110	
Unicellular greens		1547	663	110		
Total organisms per liter	225,682	154,119	103,628	63,636	44,522	
Rotifers		221	221	110		
Total organisms per liter		221	221	110		

Table 4. Vertical sampling of plankton flora and fauna (organisms per liter) taken from Lake Erie at the Crown Point Intake.

8-14-71

Organisms per liter	Surface	3.05M	6.10M	9.15M	12.20M	13.73M
Anabaena sp.		166	152	304	152	
Asterionella formosa	2279	7623	1215	8507	5772	1975
Ceratium hirundinella	2127	5303	1823	4405	9418	3646
Chroococcus sp.					304	
Closteriopsis sp.			304	304	456	152
Coelastrum microporum	152			152	152	
Coelosphaerium sp.			456	152	152	
Cosmarium sp.		166	152		152	
Dictyosphaerium sp.	456	166		304	1215	152
Dinobryon sp.	304		2734	152		
Euglena sp.	304		152			
Fragilaria crotonensis	30,382	77,888	65,321	136,718	126,388	67,751
Gleocystics sp.		663				
Kirchneriella sp.				152	152	
Micractinium sp.		331				
Mougeotia sp.	3342	17,566	12,760	21,875	36,762	16,406
Oscillatoria sp.				456	152	
Pediastrum duplex		497	304	456	152	152
Pediastrum simplex	608	994	1671	706	2734	304
Scenedesmus sp.			152		152	
Sphaerocystics sp.				2734	608	304
Spirogyra sp.		1657				
Stauroneis sp.	1823	3149	2582	5317	8811	2582
Stephanodiscus sp.						152
Tabellaria fenestrata	4557	8617	6076	6988	14,279	1215

Unicellular greens			152			
Total organisms per liter	46,486	124,786	96,006	190,192	208,419	94,791
Rotifers		166		1062	304	
Total organisms per liter		166		1062	304	

composition was observed which may have caused the problem, although Ceratium hirundinella was found at the surface in large numbers (7848 organisms per liter). It has been well documented that this species is associated with coloring of waters of reservoirs and lakes, and causes taste and odor problems in the same, although the very significant increase in the previously existing phytoplankton may have also been a major factor.

Several of the species isolated in this study, e.g., Ceratium sp., Coelosphaerium sp., Dinobryon sp., Fragillaria sp., Pediastrum sp., Staurastrum sp., and Tabillaria sp., have been reported as important taste and odor producing algae by Palmer (9, 10, 11, 12). While Mougeotia has not been reported by Palmer as an important taste and odor producing alga, its presence in large numbers in the present study suggests that it may also be involved in the production of undesirable tastes and odors.

While realizing the limitations of information gathered by means of the Hach Kit, nevertheless, the data does tend to substantiate the findings made by other workers working in the same general region.

The fungi isolated on the various media are listed in Tables 8, 9 and 10. None of the fungi isolated produced an earthy odor of any sort even though they were incubated, in most cases, for several weeks. One actinomycete culture was isolated in the August sampling and this organism did produce an earthy-musty odor.

The predominant fungi collected were various yeasts and species of Penicillium, Aspergillus and Alternaria. None of these cultures produced an odor. One isolate of Streptomyces was found and it produced an earthy-musty odor. However, as this organism was isolated but once, it is hard to visualize the actinomycetes playing any significant role in the odor problem at the Crown Plant.

An examination of the bottom mud and scrapings from the wall of the Crib revealed no taste and odor producing microorganisms although an interesting species of Phoma was isolated from material collected in the Crib.

Table 5. Vertical sampling of chemical and physical data taken from Lake Erie at the Crown Point Intake.

6-19-71

PPM	Surface	3.05M	6.10M	9.15M	12.20M	13.73M
Alkalinity ($CaCO_3$)						
Bicarbonate	80.0	90.0	86.0	100.0	99.0	
Carbonate	10.0	10.0	14.0	0.0	0.0	
Total	90.0	100.0	100.0	100.0	99.0	
Hardness ($CaCO_3$)						
Calcium	100.0	90.0	95.0	89.0	90.0	
Magnesium	21.0	37.0	30.0	30.0	30.0	
Total	121.0	127.0	125.0	119.0	120.0	
Iron	0.05	0.05	0.0	0.02	0.02	
Nitrate Nit.	0.13	0.15	0.11	0.09	0.09	
Nitrite Nit.	0.01	0.01	0.008	0.004	0.005	
Oxygen	11.0	11.5	12.0	7.0	6.5	
Ph	8.60	8.60	8.60	7.70	7.60	
Phosphate						
Ortho	0.04		0.03	0.01		
Total			0.11	0.14	0.14	
Sulfate	21.0	22.0	21.0	18.0	18.0	
Temperature (C)	21.0	20.0	19.5	15.0	11.8	
Turbidity (JT Units)	0.0	0.0	3.0	8.0	2.0	

Table 6. Vertical sampling of chemical and physical data taken from Lake Erie at the Crown Point Intake.

7-17-71

PPM	Surface	3.05M	6.10M	9.15M	12.20M	13.73M
Alkalinity ($CaCO_3$)						
Bicarbonate	65.0	65.0	62.0	64.0	68.0	67.0
Carbonate	20.0	20.0	20.0	20.0	20.0	20.0
Total	85.0	85.0	82.0	84.0	88.0	87.0
Carbon Dioxide	0.0	0.0	0.0	0.0	0.0	0.0
Hardness ($CaCO_3$)						
Calcium	93.0	94.0	95.0	94.0	92.0	98.0
Magnesium	32.0	31.0	31.0	36.0	29.0	22.0
Total	125.0	125.0	126.0	130.0	121.0	120.0
Iron	0.0	0.0	0.0	0.0	0.0	0.0
Nitrate Nit.	0.109	0.114	0.115	0.113	0.113	0.118
Nitrite Nit.	0.010	0.009	0.100	0.100	0.100	0.009
Oxygen	8.70	8.50	8.60	8.90	8.90	8.60
Ph	8.58	8.58	8.50	8.54	8.43	8.51
Phosphate						
Ortho	0.004	0.003	0.003	0.003	0.003	0.004
Total	0.005	0.005	0.004	0.005	0.004	0.005
Sulfate	20.0	18.0	20.0	20.0	19.5	19.5
Temperature (C)	21.0	21.0	21.0	21.0	20.9	21.5
Turbidity (JT Units)	0.0	0.0	0.0	0.0	0.0	0.0

Table 7. Vertical sampling of chemical and physical data taken from Lake Erie at the Crown Point Intake.

8-14-71

PPM	Surface	3.05M	6.10M	9.15M	12.20M	13.73M
Alkalinity ($CaCO_3$)						
Bicarbonate	87.0	80.0	75.0	93.0	80.0	90.0
Carbonate	10.0	20.0	20.0	10.0	10.0	0.0
Total	97.0	100.0	95.0	103.0	90.0	90.0
Carbon Dioxide	0-1	0-1	0-1	0-1	0-1	0-1
Hardness ($CaCO_3$)						
Calcium	100.0	97.0	99.0	100.0	100.0	100.0
Magnesium	25.0	33.0	22.0	30.0	20.0	30.0
Total	125.0	130.0	121.0	130.0	120.0	130.0
Iron	0.0	0.0	0.0	0.0	0.0	0.0
Nitrate Nit.	0.024	0.023	0.025	0.026	0.023	0.05
Nitrite Nit.	0.006	0.006	0.005	0.004	0.007	0.009
Oxygen	11.0	12.5	9.0	10.5	13.0	8.0
Ph	8.54	8.25	8.50	8.45	8.20	7.30
Phosphate						
Ortho	0.005	0.003	0.006	0.003	0.005	0.006
Total	0.130	0.003	0.006	0.170	0.100	0.120
Sulfate	20.0	23.0	24.0	23.0	23.0	20.5
Temperature (C)	22.0	22.0	22.7	21.2	22.6	21.2
Turbidity (JT Units)	1.0	1.0	8.0	1.0	0.0	10.0

Table 8. Vertical sampling of fungi taken from Lake Erie at the Crown Point Intake.

6-19-71

Average No. of Colonies per Petri dish	Surface	3.05M	6.10M	9.15M	12.20M	13.73M
Alternaria tenuis	13	5	2			
Penicillium sp.	5	3	1			
Aspergillus niger	8	5	3		1	
Monila sitophila	4	1		1		
Aureobasidium pullulans	10	5		3		
Sporotrichum sp.	8	2	1	1		
Saccharomyces cerevisial	2	1				
Rhodotorula	3	2	1	1	1	

Table 9. Vertical sample of fungi taken from Lake Erie at the Crown Point Intake.

7-17-71

Average No. of Colonies per Petri dish	Surface	3.05M	6.10M	9.15M	12.20M	13.73M
Aureobasidium pullulans	10	6	4	1		
Rhotorula sp.	8	7	6	2	1	1
Alternaria tenuis	3	2	1		1	
Penicillium sp.	2	1	1	1	2	1
Aspergillus niger	5	8	2	1		
Monila sitophila	2	1				
Candida sp.	4	1	1	1		1
Geotrichum candida	2	1		2	1	
Sporotrichum sp.	1	1		1		
Cladosporium sp.	1	2	1		1	
Tusarium sp.	4	1		1		

Table 10. Vertical sample of fungi taken from Lake Erie at Crown Point Intake.

8-14-71

Average No. of Colonies per Petri dish	Surface	3.05M	6.10M	9.15M	12.20M	13.73M
Aureobasidium pullulans	8	4	4	3		1
Rhodotorula sp.	10	8	7	5	4	1
Aspergillus sp.	2	1	1	2	1	
Penicillium sp.	4	2	2			
Candida sp.	3	1	2	2		1
Geotrichum sp.	2	1	2	1	1	
Alternasia tenuis	5	5	4	3		
Tusarium sp.	1	2	1	1		
Paecilomyces elegans	1	1				
Gliocladium sp.	2		1		1	
Streptomyces	1					

The information gathered from the analyses for bacteria are discussed below.

The total coliform counts are shown in Table 11. The results designated as A in the table were made at the Crown Point Intake while those designated as B in the table were taken at a sampling site one-half mile from the Crib. The designation S, as in A-S, refers to the surface sample while the number, for example A-20, refers to depth at which the collection was made. A-sd refers to the sediment sample.

Table 11. Total coliform count enumerated as organism per 100 ml.

Station	Collection Period		
	June	July	August
A-S	3650	255	10
A-20	3600	240	30
A-40	1950	253	30
A-sd	NT	700	100
B-S	16,000	4100	30
B-20	>10	310	140

In Table 12 the fecal coliforms are shown. The designation NT means not taken.

Table 12. Fecal coliform count enumerated as organisms per 100 ml.

Station	Collection Period		
	June	July	August
A-S	112	>2	>1
A-20	38	>2	>1
A-40	75 >10	>2	1
A-sd	NT	>100	>100
B-S	15	10	3
B-20	10	>2	8
B-40	NT	NT	16

The total plate counts at the various incubation temperatures are shown in Table 13. The designation NT means not taken.

In Table 14 the counts for sulfur, nitrogen and iron bacteria are expressed as well as the total yeast counts. In the portion of the table concerned with the sulfur bacteria the numbers indicate how much sample was added to the medium in order to get a positive test reaction. A positive reaction being blackening in the sulfate broth medium. If, for example, we look at station A-S for the three sampling periods, we find that a positive reaction for sulfur bacteria was noted in June and July when 10 ml of the sample was added to the medium. In August the reaction was negative for all of the samples analyzed. In the A-sd sample we find that in July there was a positive reaction when 1 ml of a 1/100 dilution of the sample was used and in August we find a positive response when 0.01 ml of a 1/100 sample was tested.

In the section of the table concerned with nitrifying bacteria the same scheme prevails; for example, in B-S we find the number +200 which means that 200 ml of the sample material was necessary in order to achieve a positive response which in this case was a change in pH.

In the section of the table dealing with iron bacteria all of the pour plates containing Waksman's medium were negative. Duplicate pour plates containing either 1.0 or 0.1 ml of sample were used. In July and August Lieoke's medium was used to enhance growth and then the organism which grew out were streaked on Waksman's medium. The asterisk denotes a positive response. A positive test being growth on Waksman's medium. In the final section of Table 14 the yeast counts per 100 ml of medium are given for July and August. This procedure was not performed in the June sample.

Table 13. Total plate counts enumerated as organisms per ml.

Station	35°			Aerobic 20°			5°			Aerobic 20°		
	June	July	August	June	July	August	June	July	August	June	July	August
A-S	2600	100	90	2200	705	825	1900	100	150	270	60	160
A-20	275	90	80	825	335	480	775	180	170	850	30	40
A-40	40	70	250	293	580	460	1100	150	140	210	50	130
A-sd	NT	14,000	219,000	NT	110,000	2690	NT	11,000	108,000	NT	10,000	34,000
B-S	625	150	370	590	685	510	1850	60	150	70	20	50
B-20	575	100	440	837	255	1130	480	50	120	180	40	30
B-40	NT	NT	740	NT	NT	3120	NT	NT	890	NT	NT	290

Table 14. Total counts for sulfur, nitrifying and iron bacteria and total yeast count.

Station	Sulfur Bacteria			Nitrifying Bacteria			Iron Bacteria			Total Yeasts/100ml		
	June	July	August	June	July	August	June	July	August	June	July	August
A-S	+10	+10	—	+10	+10	—		*	*		6	8
A-20	+10	—	—	—	+10	—			*		2	23
A-40	+10	+1	+10		+10				*			2
A-sd	NT	+1(1/100)	+0.01(1/100)			—		*	*			0
B-S	—	+10	—	+200	+10			*	*		110	12
B-20	+10	—	+10								2	7
B-40	—	NT	+1									36

SECTION VI

DISCUSSION

The present investigation was concerned primarily with a microbiological survey of the Crown Point Station. The three major groups of microorganisms studied were the fungi, bacteria and algae. The results of this study show that the fungi and bacteria play little if any role in the taste and odor problem present at the Crown Station Inlet. While a fair diversity of both fungi and bacteria were noted, the total counts were not abnormally high nor did individual cultures exhibit any pronounced odors.

Significant progress was made in identifying and enumerating the species of the algal community associated with the Crown Point Intake. Many of the species observed have been associated with taste and odor problems in other aquatic environments. During the period of this study no pronounced taste and odor difficulties were experienced at the Crown Plant. However, various local people remarked during the August sampling period that the water had what they described as a typical "Lake Erie odor". Whatever the source of these odors, they were not due to benthic or periphyton algae, but they could have been associated with the phytoplankton community within the area as the reported "Lake Erie odor" coincided with the increase in phytoplankton.

From the results of this survey, it is apparent that a continuation of the study should be continued. Much more intensive work is needed in the field to more rigorously characterize the physical, chemical and biological relationships of the area, through which patterns can be developed and models for prediction of cause and effect relationships of taste and odor problems in Lake Erie. These prediction models will enable the filtration plants to prepare for the problems and counter them when they arise.

Intensive laboratory work is needed to culture species of the phytoplankton community found around the Crown Point Intake. This culturing should be done with the natural chemical and physical environment in mind. Thus, stimuli of phytoplankton growth can be detected, especially taste and odor species, and controls may be found. Culturing should also be used to identify chemical by-products of algae which cause the taste and odor problems and possible chemical controls may be applied.

SECTION VII

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SECTION VIII

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SECTION IX

GLOSSARY

Actinomycetes - Filamentous bacteria.

Algae - Chlorophyll-containing plants lacking roots, stems or leaves.

Fungi - Chlorophyll-lacking plants which have no roots, stems or leaves.

Chrysophyta - A group of algae characterized by the formation of a yellow-brown pigment.

Chlorophyta - A group of algae characterized by the formation of green coloring materials primarily chlorophyll.

Cyanophyta - A group of algae characterized by blue-green coloring materials.

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27 Abstract

The taste and odor constituents produced by the actinomycete Streptomyces odorifer, the alga Synura petersenii and the mold Trichoderma viride were examined.

The odorous constituents were obtained by steam distillation of the culture medium. The odorous constituend were identified by means of gas-chromatography, infrared, mass and nuclear magnetic spectroscopy. The major odorous constituend produced by the above named organisms have been enumerated. (Collins-U.Conn.)

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