

**SPECIAL REPORT NO. 8**

**Studies on the Effects of Dredged Materials  
from Selected Great Lakes Harbors  
on Plankton and Benthos<sup>†</sup>**

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## INTRODUCTION

Within the past ten years we have become increasingly aware of changes taking place in the environment and biota of the Great Lakes, many of which are attributed to pollution of the lakes (Beeton, in press). The major harbors of the lakes are among the most polluted environments in the system and concern has developed about the dumping of sediments dredged from these harbors in the open lake. Consequently, the U.S. Army Corps of Engineers undertook a study, with the cooperation of the Federal Water Pollution Control Administration, to obtain acceptable solutions to the problem of the disposal of polluted dredgings (U.S. Army, Corps of Engineers, 1969).

Little information was available on the biological, chemical, and physical characteristics of the harbor and lake disposal environments. Almost ninety years ago, Vorce (1882) became concerned about changes in the abundance and species composition of plankton in the Cleveland, Ohio, water supply just two years after the city began using Lake Erie as a regular dumping ground. Oddly enough, no research has been conducted on the direct effects of spoil disposal on plankton or any other organisms in the Great Lakes. Recent studies on spoil disposal in Chesapeake Bay showed temporary increases in turbidity and nutrients in the vicinity of the disposal area. In conjunction with the Chesapeake Bay study, Flemer et al. (1968) conducted a field study on the biological effects of spoil disposal. They concluded that field studies cannot adequately evaluate the effects of sediment disposal on biota, and that suitable laboratory studies are needed.

In the Great Lakes dredging and water quality project, field investigations were undertaken to determine: (1) the nature of the environments, (2) the effects of dredging on the harbors, and (3) the effects of disposal on the lake environments (U.S. Army, Corps of Engineers, 1969). It soon became apparent that the results of

these studies would not adequately describe the effects on the biota of open-lake disposal. Consequently, laboratory studies were made at the University of Wisconsin of the physical and chemical characteristics, treatability (aerobic stabilization, anaerobic digestion, chlorine demand, calorimetry), and effect on the biota of sediments from the harbors of Buffalo, N. Y. ; Calumet, Ill. ; Cleveland, Ohio; Great Sodus Bay, N. Y. ; Green Bay, Wis. ; Indiana Harbor, Ind. ; Milwaukee, Wis. ; Rouge River (Detroit, Mich. ); and Toledo, Ohio (see Figure 6).

Functional tests of the physical and chemical characteristics, and treatability of the sediments were made in the Sanitary Engineering Laboratory, University of Wisconsin-Madison. Results of these tests are presented in the U. S. Army, Corps of Engineers (1969) report (Volume 7, Appendix C5).

This report presents the methods which were developed and the results of biological tests which were made at the Center for Great Lakes Studies, University of Wisconsin-Milwaukee. Studies were conducted using benthic organisms (amphipods and midge larvae), the benthic alga, Cladophora sp. , phytoplankton, unialgal cultures, zooplankton and Daphnia pulex in culture. The tests are simple in design and straight-forward in approach. Simplicity of experimental design was desirable because of the large number of samples to be analyzed.

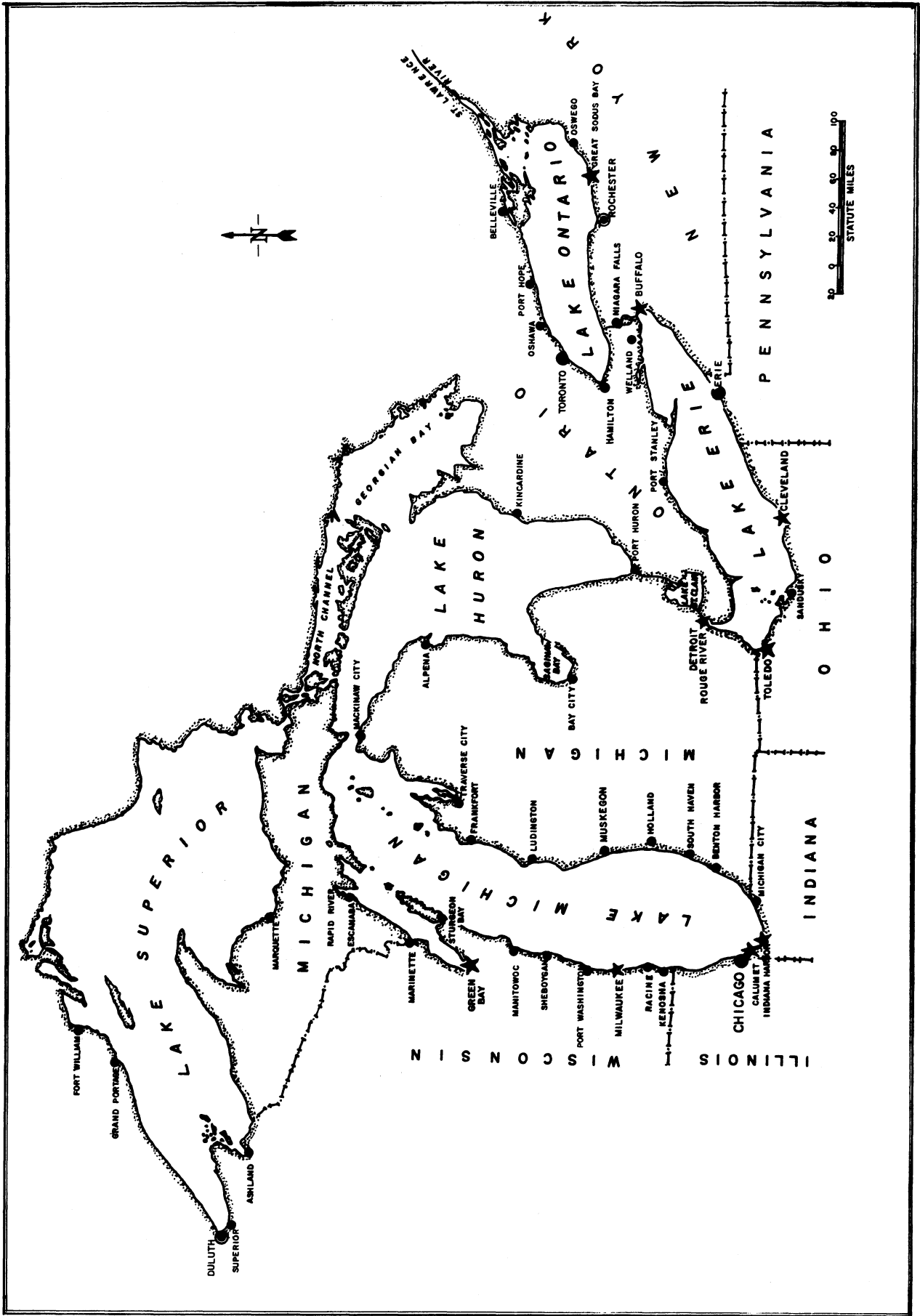


Fig. 1. The Great Lakes region, with harbors sampled in this study marked by a star.

## II. SAMPLING LOCATIONS AND SEDIMENT CHARACTERISTICS

Samples of surface sediments were obtained from five locations: Buffalo, Calumet, Cleveland, Green Bay, Indiana, Rouge River and Toledo harbors (Figs.2-8). Three samples were obtained from Great Sodus Bay (Fig. 9), one from Milwaukee harbor (Fig. 10) and one from the bin of the hopper dredge when the ship was operating in the turning basin of the Rouge River. Calumet, Green Bay, Indiana, Milwaukee, Rouge River and Toledo harbors were sampled by personnel from the Center for Great Lakes Studies (Table 1). The State University College at Buffalo, N. Y., collected samples from Buffalo, Cleveland, and Great Sodus Bay. Two gallons of sediment were collected at each location with Ekman grabs, except at Green Bay, where a Ponar grab was used. Samples were kept on ice during transit and held in a cold room until needed for the tests. Half of each sample was used for the functional tests at the Sanitary Engineering Laboratory, University of Wisconsin-Madison (U. S. Army, Corps of Engineers, 1969) and the remainder was used for the biological tests at Milwaukee. At station S-3 of Great Sodus Bay, it was not possible to collect two gallons of sediment since the bottom consisted of compacted sand and gravel. Consequently, only one gallon of sediment was obtained which was used for functional testing only.

All samples collected seemed representative of the harbor environments except Green Bay. Dredging operations preceded the sampling program at that harbor only. Sediments obtained for testing from Green Bay probably are not representative of prevalent conditions at that harbor.

TABLE 1. — Sampling Locations and Dates when Samples were Obtained.

Harbor	Date Sampled	Organization
Buffalo, N. Y.	14 June 1968	SUCB*
Calumet Harbor, Ill.	14 May 1968	CGLS†
Cleveland, Ohio	24 June 1968	SUCB
Great Sodus Bay, N. Y.	15 June 1968	SUCB
Green Bay, Wis.	15 May 1968	CGLS
Indiana Harbor, Ind.	15 May 1968	CGLS
Milwaukee, Wis.	6 May 1968	CGLS
Rouge River (Detroit, Mich.)	17 May 1968	CGLS
Rouge River‡ (Detroit, Mich.)	16 July 1968	COE§
Toledo, Ohio	16 May 1968	CGLS

\*SUCB: State University College at Buffalo

†CGLS: Center for Great Lakes Studies

‡Obtained from the bin of the hopper dredge while operating in the Rouge River.

§COE: U. S. Army, Corps of Engineers

Physical characteristics of the sediments are presented in Table 2, along with the designation of sampling locations that are used throughout the report. Some additional materials were used in the biological tests. Fuller's Earth, open-lake sediment obtained about 7 miles northeast of Milwaukee harbor, and clean aquarium sand were used as controls.

Some chemical characteristics of the sediments are presented in Table 3. These parameters will be used in interpretation of some of the biological results throughout this paper. Additional physical-chemical data including particle size analysis, pH, total solids, per cent water, specific resistance, and wet and dry density were also collected (U. S. Army, Corps of Engineers, 1969).

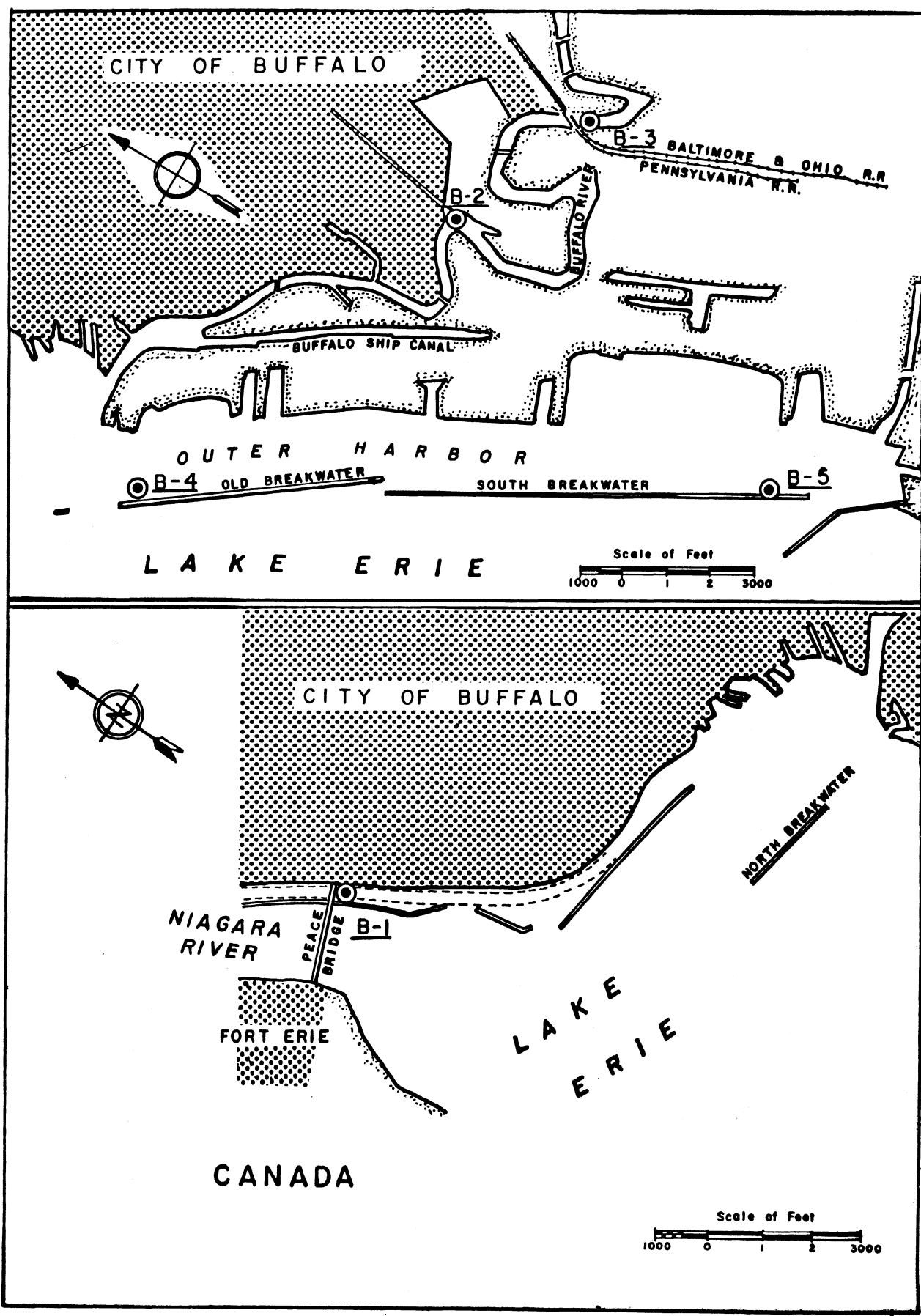


Fig. 2. Location of sampling points, 1968, Buffalo harbor.

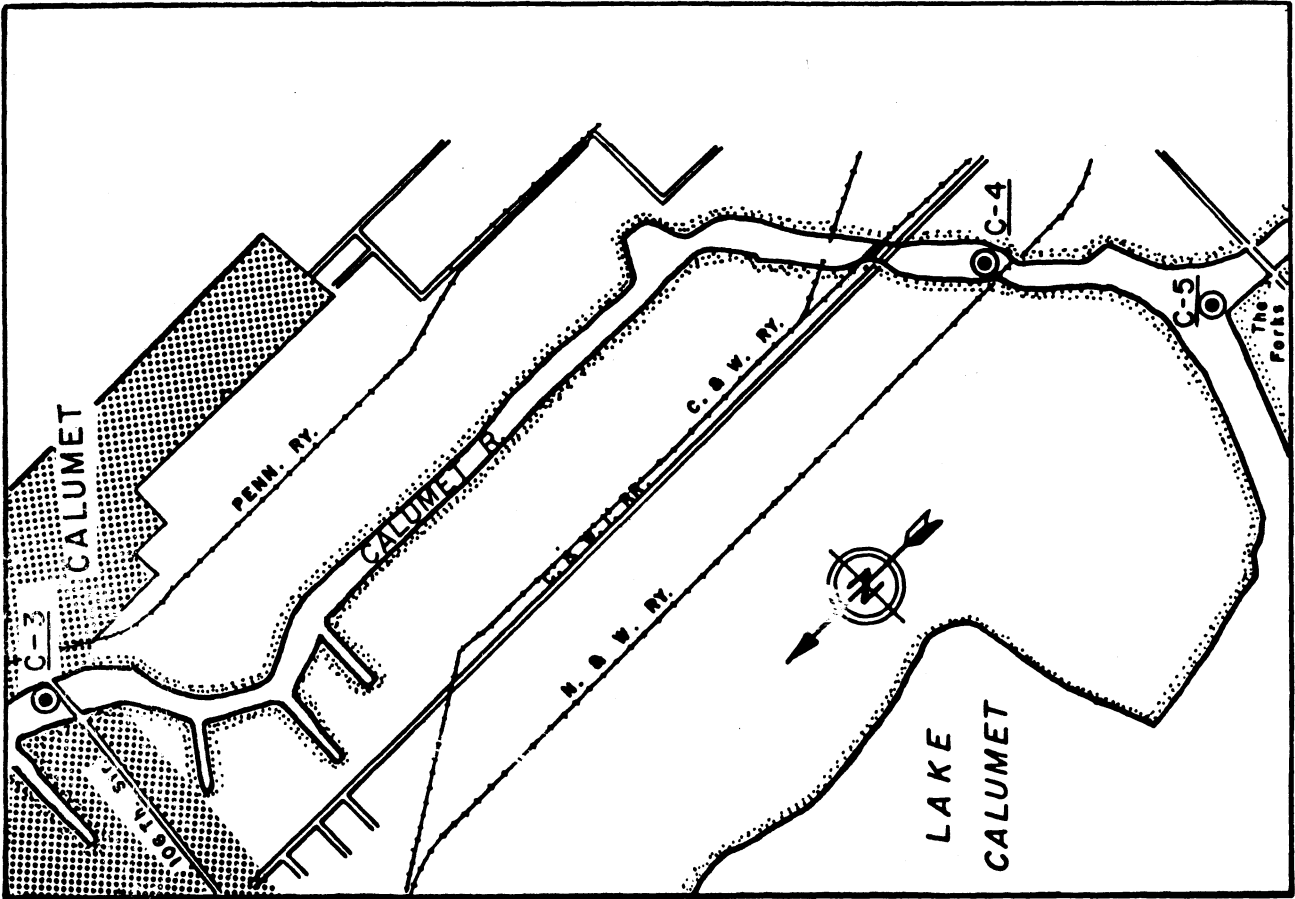
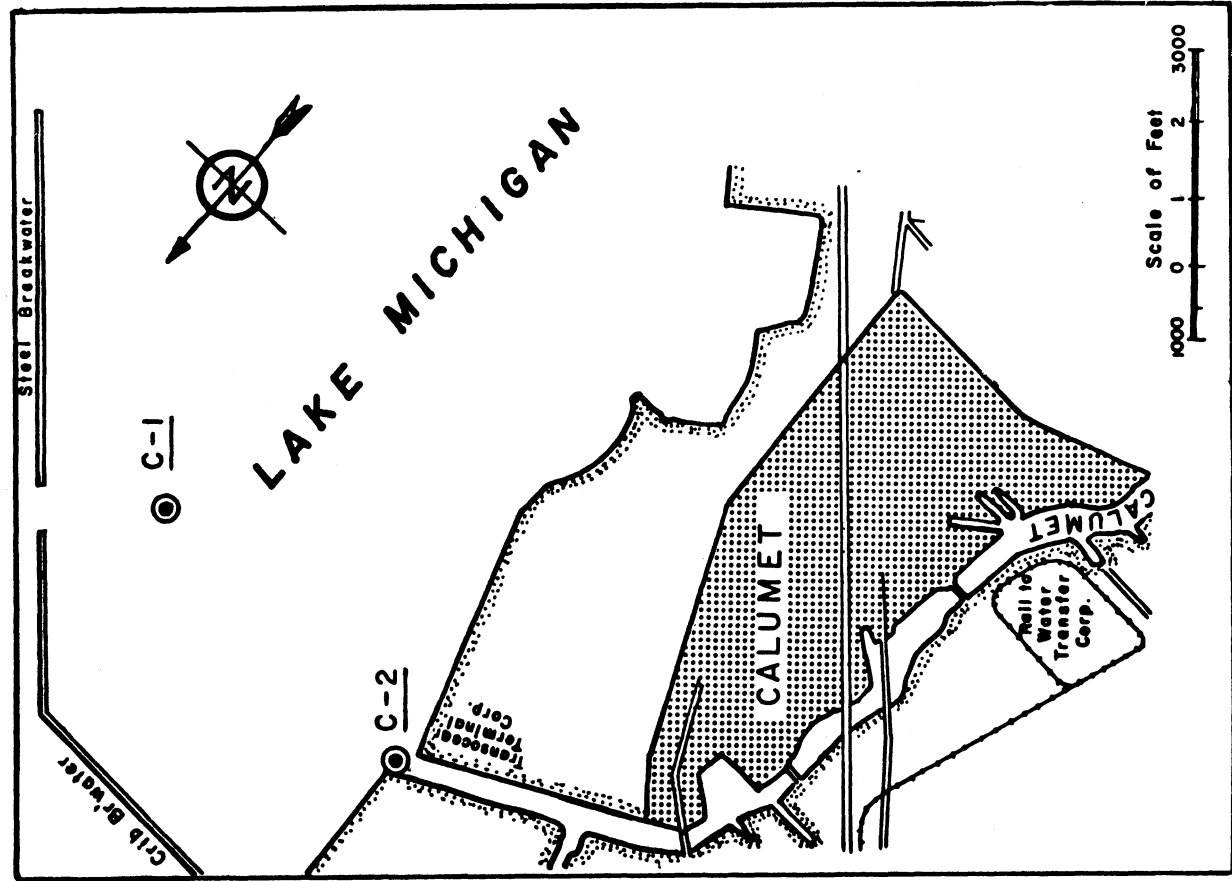


Fig. 3. Location of sampling points, 1968, Calumet river.

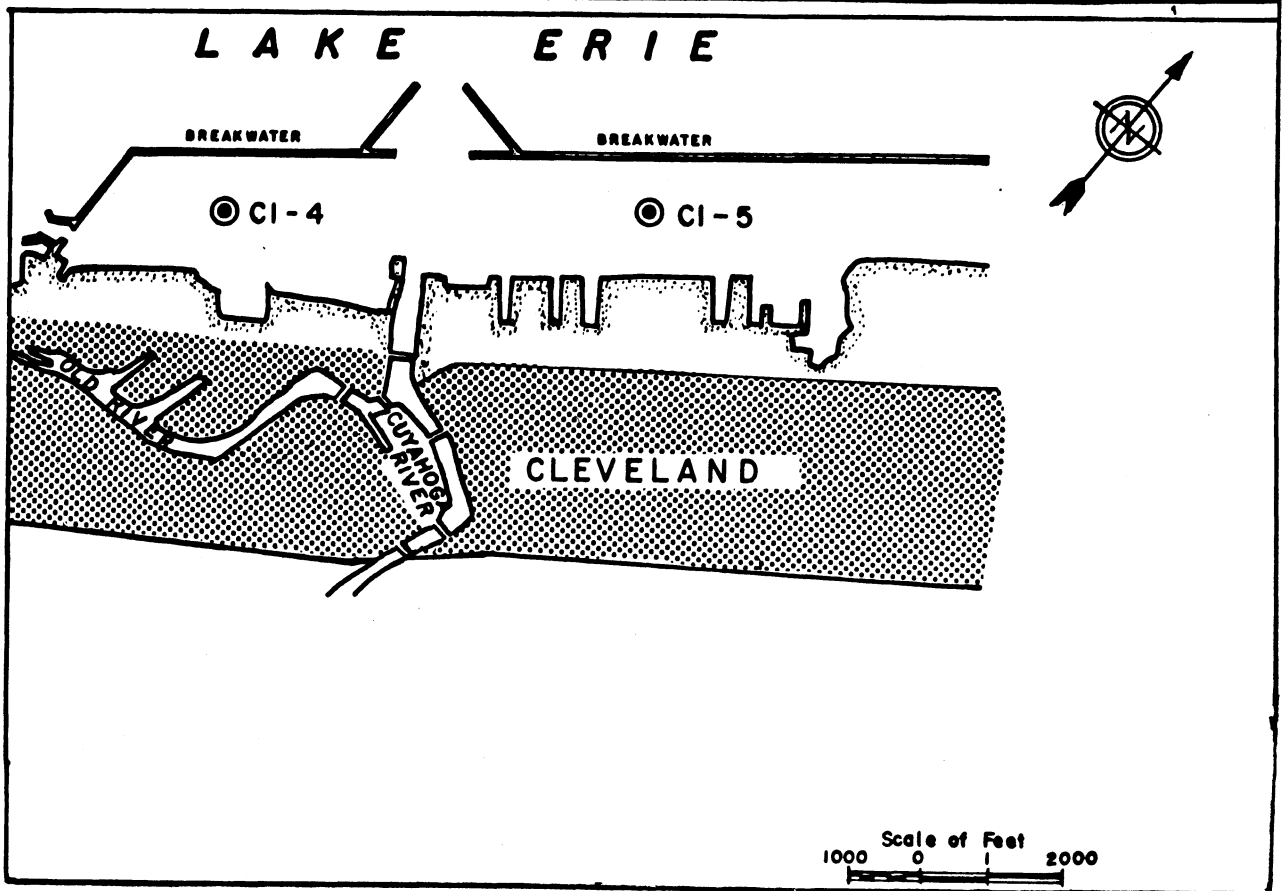
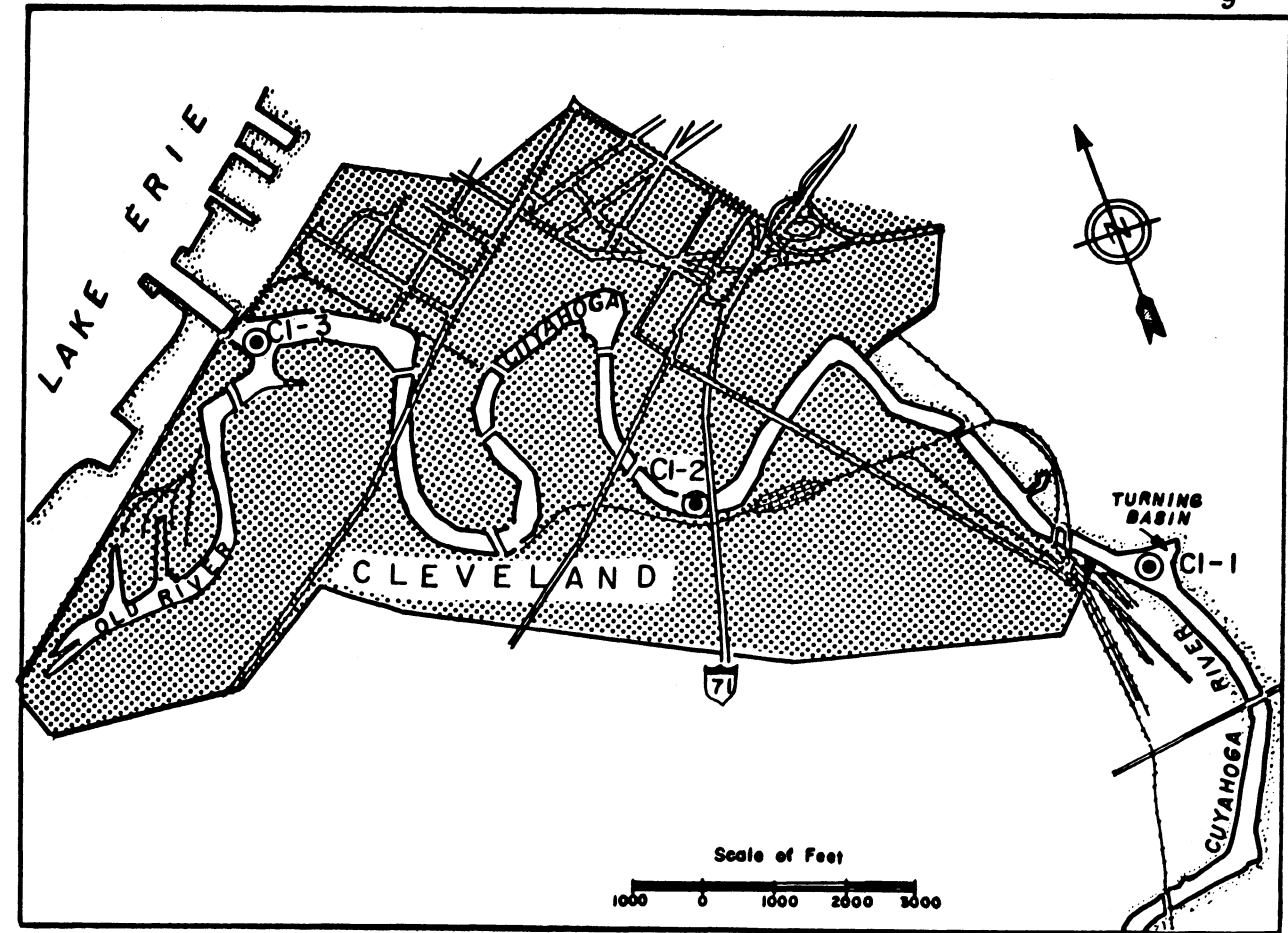


Fig. 4. Location of sampling points, 1968, Cleveland.

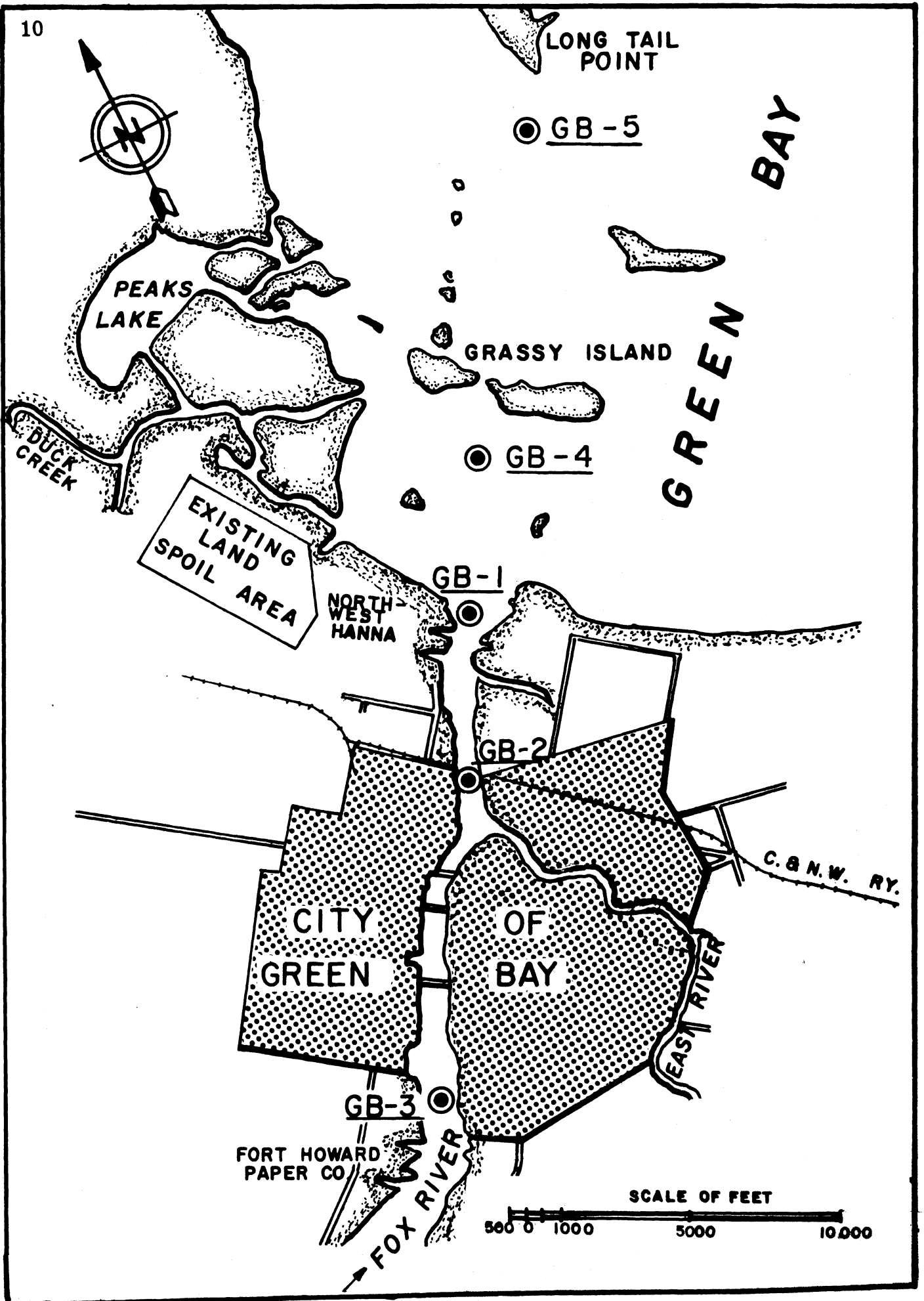


Fig. 5. Location of sampling points, 1968, Green Bay.

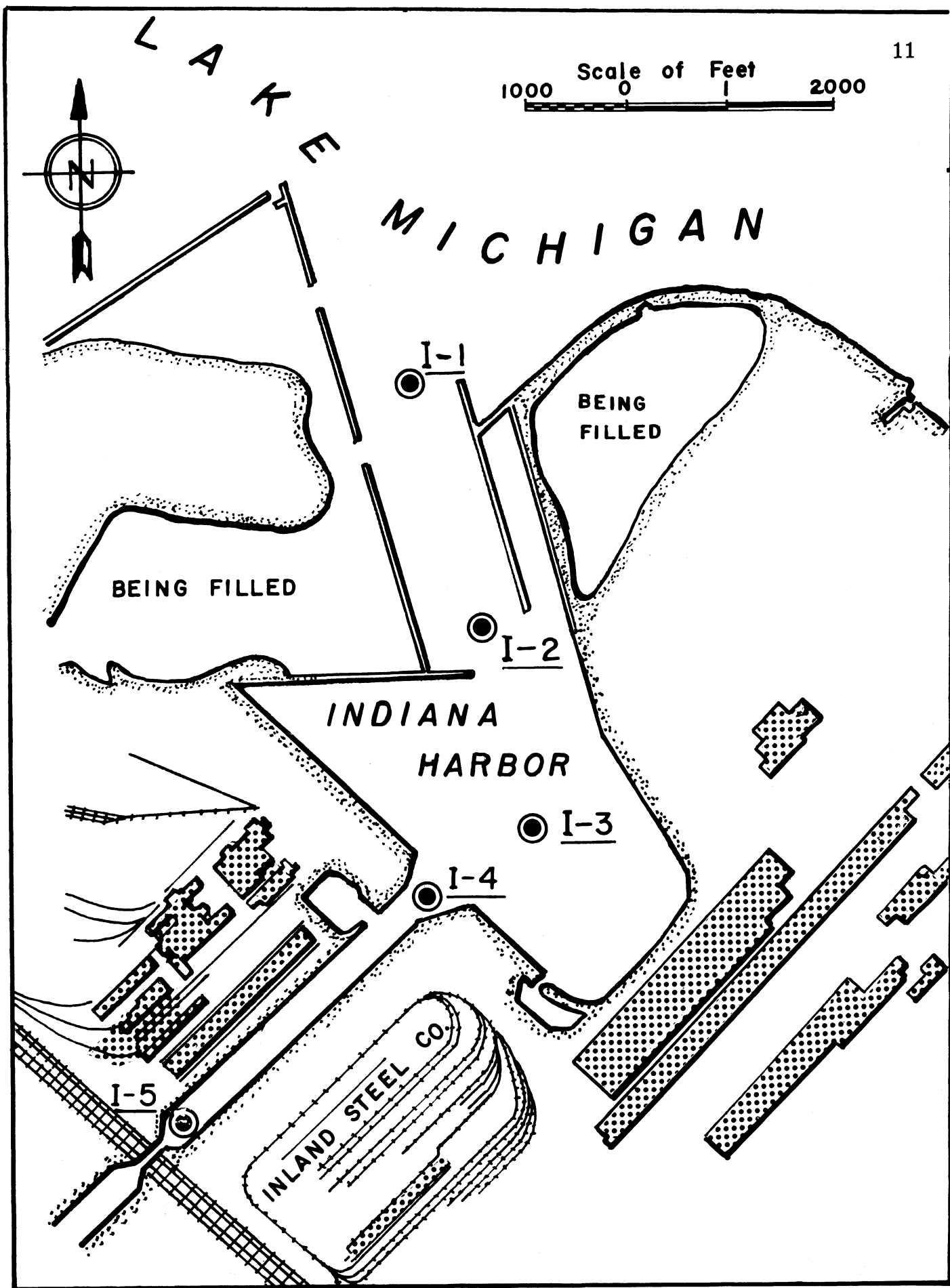


Fig. 6. Location of sampling points, 1968, Indiana harbor.

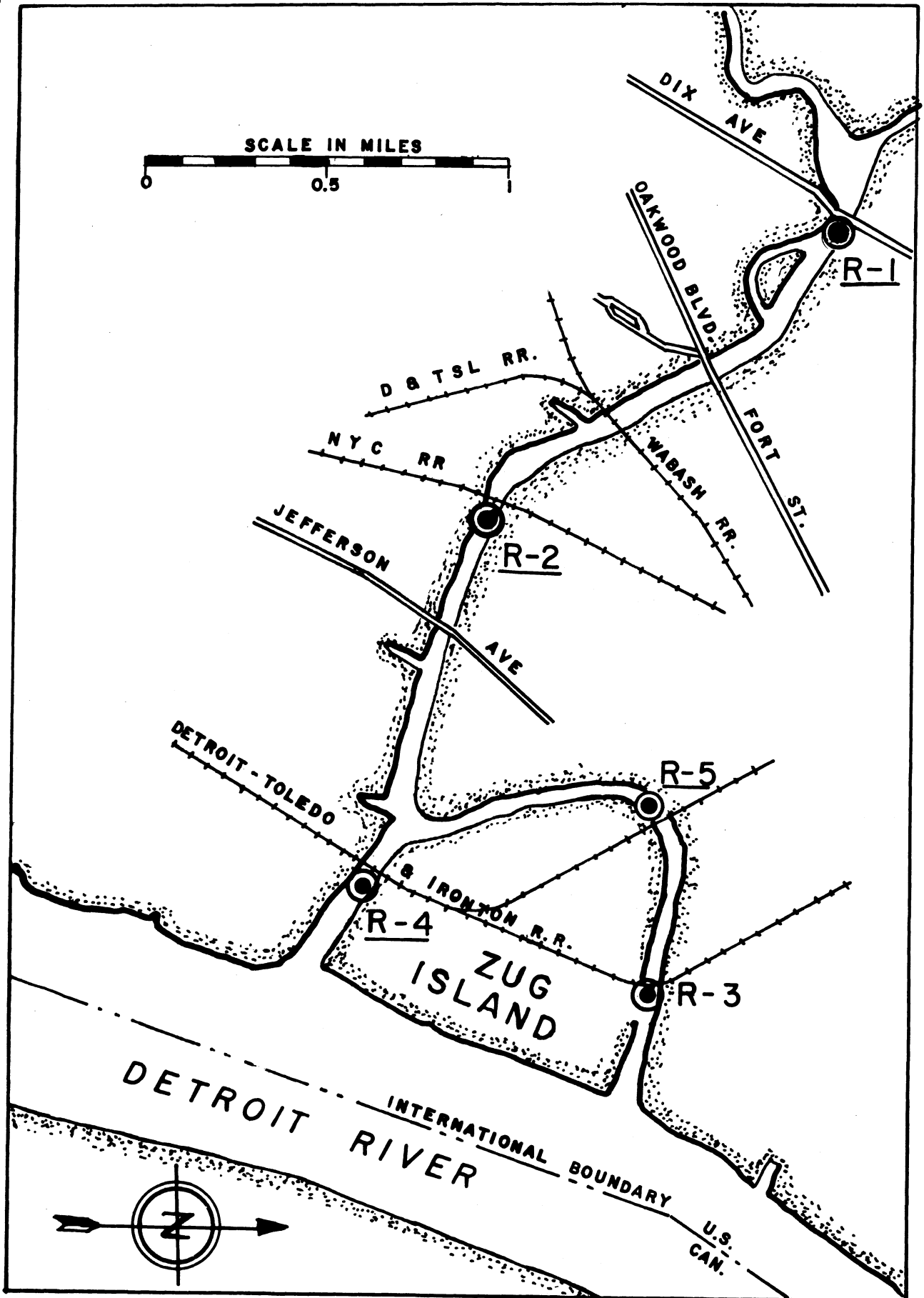


Fig. 7. Location of sampling points, 1968, Rouge River . . .

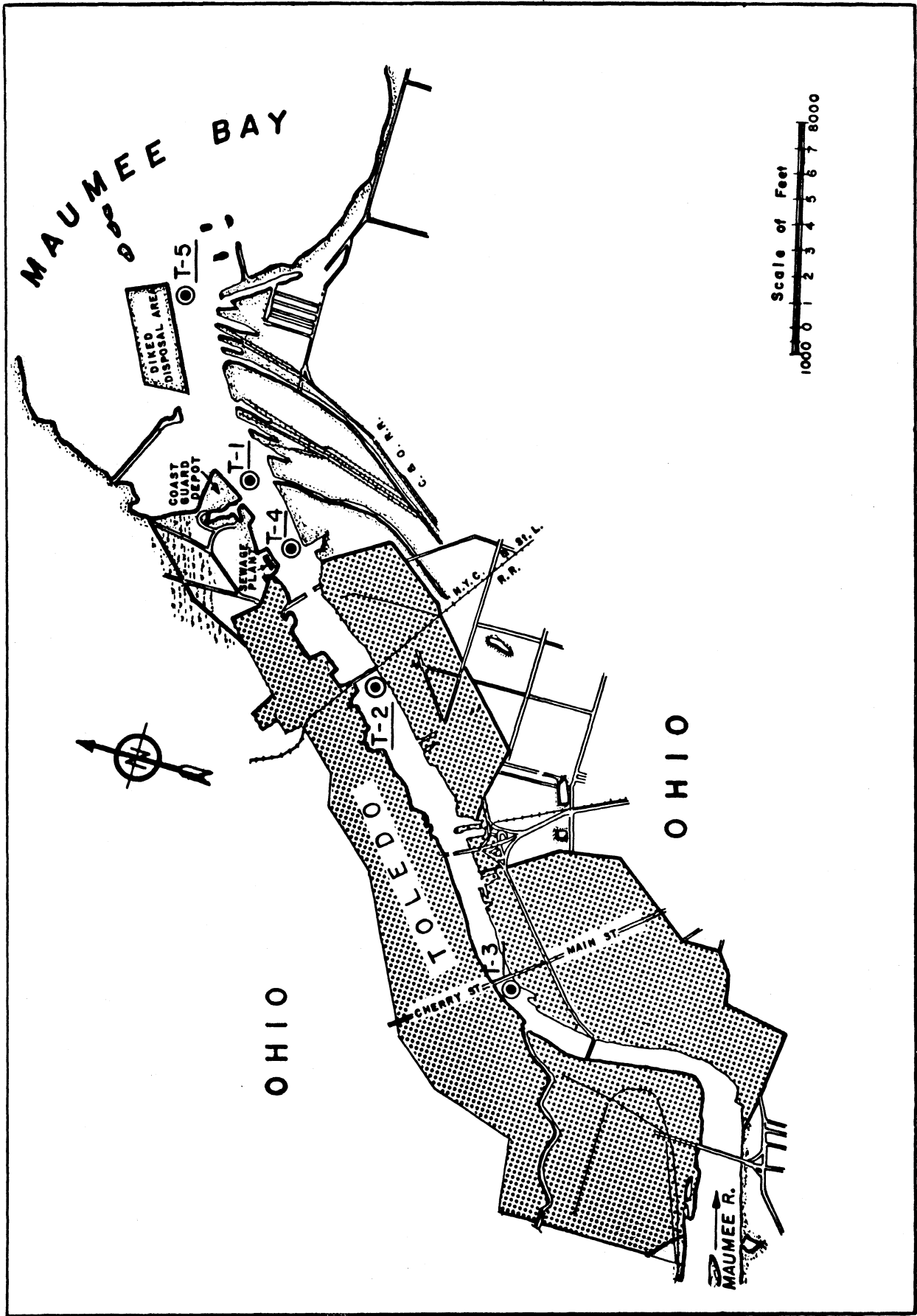


Fig. 8. Location of sampling points, 1968, Maumee River (Toledo).

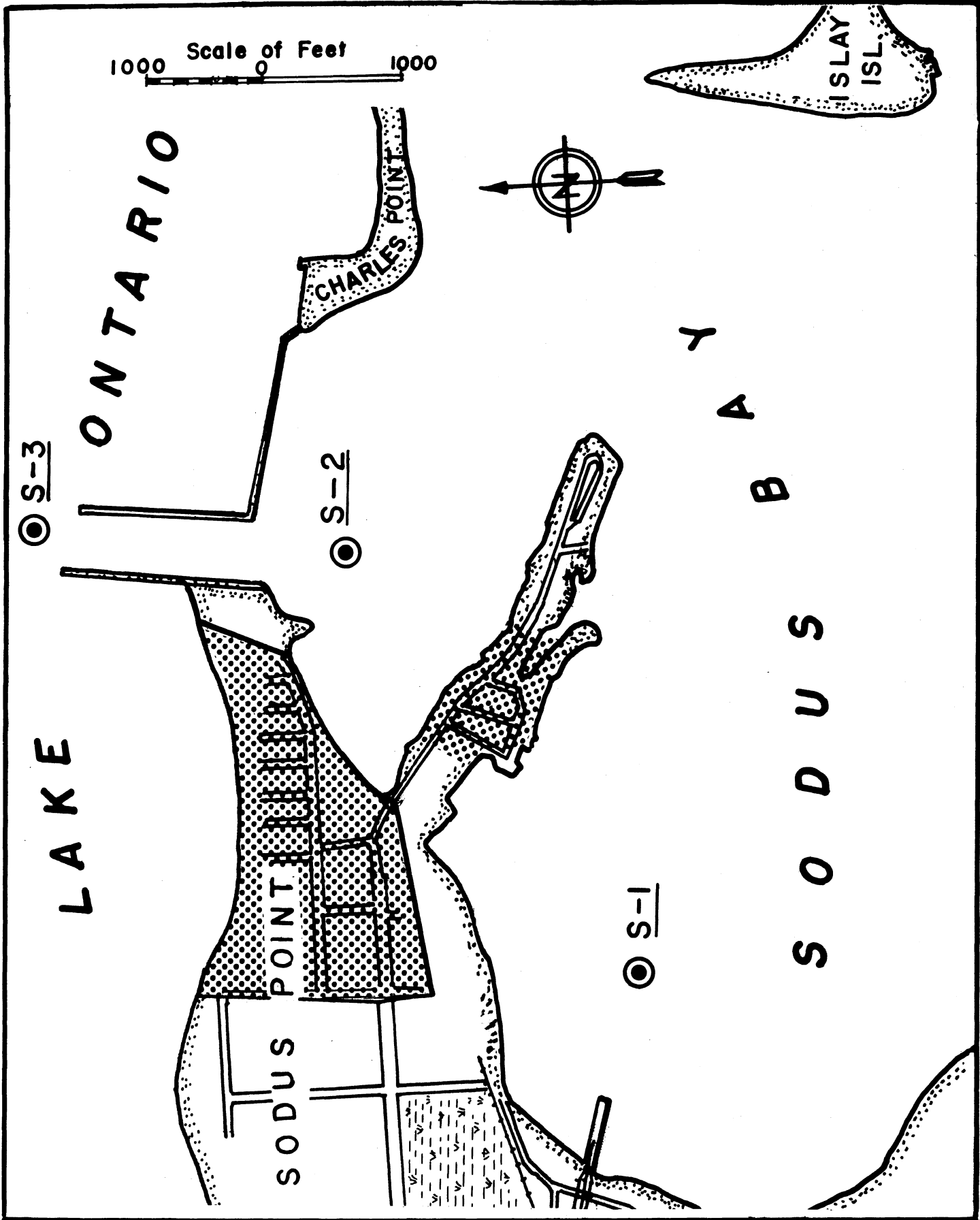


Fig. 9. Location of sampling points, 1968, Great Sodus Bay.

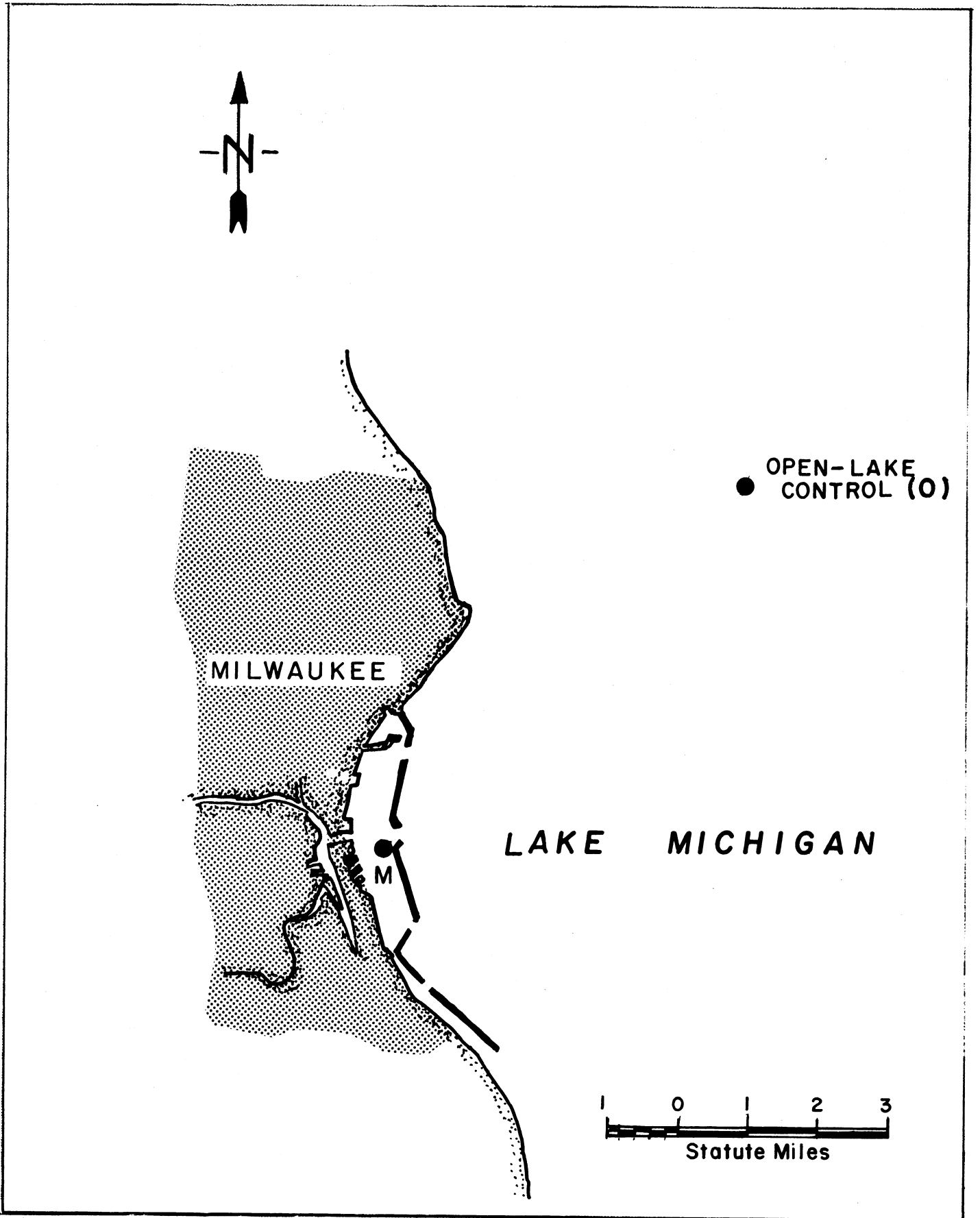


Fig. 10. Location of sampling points, 1968, Milwaukee harbor.

TABLE 2. -- Physical characteristics and observations of harbor sediment samples.

Station	Particle Size Analysis				Gross Observations of Mud Samples
	% Gravel	% Sand	% Silt	% Clay	
<b>Buffalo</b>					
B-1	0	0.9	83.0	15.9	Putrid, gray-black mud with oil, some worm trails
B-2	0.3	32.1	56.6	10.9	Black mud with oil sludge, some worm trails
B-3	0	51.4	45.4	3.4	Brown-red mud, with oil sludge, some worm trails
B-4	0.2	2.9	87.0	9.7	Putrid, brown mud
B-5	0	4.9	81.0	14.2	Putrid, brown mud
<b>Calumet</b>					
C-1	0	52.2	40.0	11.2	Muddy sand, no worm activity, no oil
C-2	0.7	12.8	69.8	16.7	Putrid reddish-brown mud, oil
C-3	0	13.0	58.8	28.2	Putrid reddish-brown mud, little worm activity, oil
C-4	0.5	25.4	34.5	39.6	Putrid gray mud, many worm trails, little oil
C-5	0	0.8	39.5	59.7	Putrid gray mud, many worm trails, little oil
<b>Cleveland</b>					
CL-1	0.2	8.9	83.5	7.4	Black mud, slightly putrid and oily smell
CL-2	6.3	9.9	75.0	8.5	Gray-black mud, slightly putrid and oily smell
CL-3	0.3	10.3	83.4	6.1	Gray-black mud, putrid, oil
CL-4	0	6.1	64.5	29.4	Gray mud, putrid, oil
CL-5	2.6	10.4	55.3	31.5	Gray silty mud, slightly putrid, little oil

Green Bay									
GB-1	0.3	43.7	24.9	31.2	Gray mud and hard clay, some worm activity, putrid odor, no oil				
GB-2	45.6	36.2	9.2	8.9	Gray mud, hard clay and considerable gravel, some worm activity, putrid odor, no oil				
GB-3	0	54.8	21.5	23.8	Gray mud and hard clay, some worm activity, putrid odor, no oil				
GB-4	0	0.8	44.5	54.7	Gray mud and hard clay, some worm activity, putrid odor, no oil				
GB-5	7.2	9.9	30.9	52.0	Pink clay, not putrid, no noticeable bioactivity				
Indiana									
I-1	0.2	47.7	41.0	11.0	Gray mud, some worm activity, oil				
I-2	2.6	27.7	62.6	7.0	Gray mud, no worm activity, smells strongly of oil				
I-3	1.1	20.2	71.0	8.7	Reddish-black mud, putrid, no worm activity, oil				
I-4	0	17.9	70.2	11.9	Reddish-brown mud and oil, no worm activity				
I-5	5.8	33.5	50.2	10.5	Reddish-brown mud, smells of oil, no worm activity				
Milwaukee									
M	not performed				Black, putrid mud from harbor entrance, some worm activity				
O	not performed				Open-lake muddy sand, some worms, midge larvae and many amphipods				
R-1	2.1	42.4	20.2	35.4	Gray mud, very putrid, tremendous numbers of worms, oil				
R-2	2.4	15.0	56.8	25.8	Gray mud, very putrid, tremendous numbers of worms, heavy oil				
R-3	1.0	50.0	33.3	15.6	Gray mud and cinders, very putrid, tremendous numbers of worms, oil				
R-4	2.6	43.0	36.3	15.3	Gray mud and much organic debris, very putrid, tremendous numbers of worms, little oil				
R-5	1.3	54.4	33.2	11.2	Gray mud and much organic debris, very putrid, tremendous numbers of worms, little oil				

Table 2, continued

Station	Particle Size Analysis			Gross Observations of Mud Samples	
	% Gravel	% Sand	% Silt %Clay		
Hopper Dredge (Rouge River)					
RH	not	performed		Black, putrid, oily mud with much organic debris	
Great Sodus Bay					
S-1	0	10.6	73.0	16.6	Putrid, brown mud, no oil
S-2	0	62.0	32.6	5.3	Brown muddy sand, worm trails present, no oil
Toledo					
T-1	0	1.8	37.0	61.4	Gray mud, putrid, some worm activity, no or little oil
T-2	0	2.2	40.4	57.4	Gray mud, putrid, some worm activity, no or little oil
T-3	0.6	21.4	39.9	38.0	Gray mud, putrid, some worm activity, no or little oil
T-4	0	10.2	71.6	18.2	Gray mud, putrid, some worm activity, no or little oil
T-5	3.3	18.7	47.2	30.8	Gray mud, putrid, some worm activity, no or little oil
Fuller's Earth					
F	not	performed			Technical grade diatomaceous earth
Control					
C	not	performed			Clean, coarse aquarium sand and fine gravel

TABLE 3. — Some chemical characteristics of the harbor sediments.<sup>†</sup>  
 (COD = chemical oxygen demand; NH<sub>3</sub>-N = ammonia-nitrogen;  
 org-N = organic nitrogen; and PO<sub>4</sub>-P = phosphate phosphorus).

Station	COD mg/liter	NH <sub>3</sub> -N mg/gm	Org-N mg/gm	PO <sub>4</sub> -P mg/gm	Volatile so- lids mg/liter
Buffalo					
B-1	23,250	0.058	1.97	0.66	21,040
B-2	53,250	0.071	2.06	0.53	38,500
B-3	66,250	0.056	1.98	0.79	46,417
B-4	53,500	0.098	2.55	0.51	37,350
B-5	43,600	0.050	1.43	0.48	30,420
Calumet					
C-1	21,700	0.052	1.58	0.41	14,370
C-2	64,000	0.035	0.76	0.44	41,930
C-3	69,200	0.066	1.45	0.64	48,730
C-4	63,700	0.050	1.25	0.54	49,550
C-5	41,600	0.044	1.14	0.39	41,910
Cleveland					
CL-1	141,500	0.146	2.04	1.02	61,780
CL-2	91,200	0.124	1.49	0.72	52,090
CL-3	101,500	0.123	1.90	0.98	55,360
CL-4	52,650	0.106	1.98	0.68	42,250
CL-5	44,900	0.179	1.77	1.02	46,070
Green Bay					
GB-1	48,400	0.091	4.34	0.79	47,330
GB-2	42,300	0.068	1.40	0.51	45,270
GB-3	40,000	0.091	3.23	0.98	39,320
GB-4	38,300	0.410	3.92	0.83	38,370
GB-5	17,600	0.050	4.47	0.35	23,230
Indiana					
I-1	70,100	0.085	1.68	0.48	48,610
I-2	60,300	0.059	1.44	0.72	52,440
I-3	77,300	0.033	1.40	0.46	36,340
I-4	141,500	0.072	2.98	1.05	66,460
I-5	217,500	0.052	2.20	0.86	75,950

<sup>†</sup>No chemical analyses were made on the Milwaukee harbor samples.

Table 3, continued

Station	COD mg/liter	NH <sub>3</sub> -N mg/gm	Org-N mg/gm	PO <sub>4</sub> -P mg/gm	Volatile so- lids mg/liter
<b>Rouge River</b>					
R-1	142,000	0.091	2.08	0.89	64,260
R-2	83,900	0.094	2.20	0.96	65,160
R-3	46,900	0.062	3.60	0.72	57,730
R-4	142,600	0.052	2.34	0.74	79,240
R-5	106,000	0.063	2.20	0.57	60,380
<b>Hopper Dredge</b>					
RH	28,800	0.051	1.98	0.81	24,800
<b>Sodus Bay</b>					
S-1	44,000	0.057	2.38	0.46	33,450
S-2	22,150	0.053	2.00	0.33	19,900
<b>Toledo</b>					
T-1	35,300	0.128	3.35	0.72	38,520
T-2	41,800	0.125	1.81	0.81	44,830
T-3	30,400	0.081	1.77	0.70	54,990
T-4	47,100	0.235	3.80	0.99	47,020
T-5	31,100	0.078	1.90	0.58	42,750

### III. EXPERIMENTAL METHODS AND PROCEDURES

#### A. Selection of Sediments by Benthic Organisms

One quart capacity milk cartons were cut off  $2\frac{1}{2}$  in from the bottom, and this bottom section was completely filled with the test sediment. The cartons were then placed side-by-side in a 30-gallon aquarium according to a table of random numbers. The cartons were covered with cheesecloth and the aquarium was slowly filled to prevent disturbance of the sediments. The cheesecloth was removed and the test animals, amphipods and midge larvae, were introduced randomly over the water surface. The deepwater amphipod, Pontoporeia affinis, collected in Lake Michigan 7 mi northeast of Milwaukee harbor, and a stream form, Gammarus lacustris, collected near Eagle, Waukesha County, Wisconsin, were used in the first set of experiments. Midge larvae, Chironomus tentans, obtained from the Root River, Milwaukee County, Wisconsin, were used in the second set of experiments.

The water temperature in the aquarium was maintained at about 12.5 C for the amphipod experiments and at about 20 C for the midge larvae experiments. The amphipod tests ran 60 hrs and the midge larvae 48 hrs. Each experiment was run in duplicate. Cheesecloth was placed over the sediments at termination of the tests and the water was quickly drained. The contents of each carton were screened through a No. 35 sieve, and the living and dead organisms were counted immediately.

## B. Benthos Viability Studies

The amphipods, Pontoporeia affinis and Gammarus lacustris, and the midge larvae, Chironomus tentans, were used for these tests also. The organisms were collected just prior to experimentation and screened out of their native sediment with a No. 30 sieve. In the amphipod experiments, six organisms were placed in 8 oz wide-mouth jars, which contained 1–2 in of test sediment and aerated Lake Michigan water. The tops of the jars were covered with cheesecloth, secured by a rubber band, and placed in a 30-gallon aquarium. Placement in the tank was determined by a table of random numbers. Lake Michigan water (dechlorinated city water) was circulated vigorously through the tank to maintain the high dissolved oxygen content and a low temperature (about 12.5 C). One experiment was terminated in 24 hours, the other in 48 hrs, and the contents of each jar were washed through a No. 35 sieve to recover living and dead organisms. The test procedure was the same for midge larvae except that 60 ml bottles were filled to about 1.3 full with test sediment and the temperature was maintained at about 18 C.

## C. Bioassay of Sediments Using Phytoplankton and Algal Culture

### 1. Phytoplankton

Only small amounts of sediment were used in these tests. A level teaspoon of sediment was placed in 500 ml of distilled water and stirred at a uniform speed on a magnetic stirrer. One, 2.5, 5, and 10 ml samples were withdrawn with pipettes while the diluted sediment was stirred. These volumes of sediment were added to 50 ml of phytoplankton in a 60 ml bottle. The phytoplankton consisted primarily of the blue-green alga Oscillatoria rubescens and some Anabaena sp. and Gomphosphaeria sp., from the Juneau Park Lagoon, Milwaukee, Wisconsin.

A control, i. e. , phytoplankton in 60 ml bottles without sediment, was conducted along with each harbor sediment. Changes in the number of phytoplankton cells, which should be a measure of growth or decline of the population, was measured by determining the optical density with a Bausch and Lomb Spectronic 20 photometer set at wavelength 525 m $\mu$ . A measurement was made as soon as the diluted sediment was added to the phytoplankton and every 24 hrs thereafter . Temperature was maintained at about 18 C. The bottles were vigorously shaken 4 to 6 times daily and immediately before each measurement. Experiments were terminated in 7 to 10 days.

## 2. Algal cultures

The algal cultures were subjected to the same procedure as above except that the known volumes of diluted sediments were added to 50 ml of Chlorella pyrenoidosa<sup>1</sup> and 50 ml of Anabaena flos-aquae<sup>2</sup> , both in Gorham's culture medium (Hughes, Gorham, and Zehnder, 1958).

### D. Bioassay of Sediments Using Carbon-14 Uptake by Phytoplankton

A carbon-14 bioassay method modified from Wetzel (1965a) was developed. Sediment extracts were prepared to avoid the possibility of adsorption of carbon-14 on sediment particles. Fifty ml of sediment was added to 250 ml of distilled water in a 600 ml beaker and stirred for 10 minutes on a magnetic stirrer. The slurry was poured into a 500 ml flask, allowed to settle for 24 hrs, then shaken vigorously. This settling and shaking procedure was continued every 24 hrs for 4 days. On the fifth day the supernatant was poured through No. 1

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<sup>1</sup> University of Wisconsin-Madison culture 2005.

<sup>2</sup> Indiana University culture 1444.

Whatman's filter paper. The filtrate was then poured through No. 2 Whatman's filter paper. The final filtrate was refrigerated until used in the tests.

One mc of  $\text{NaC}^{14}\text{O}_3$  was mixed with 25 liters of water ( $40 \mu\text{c}/\text{liter}$ ), which had been collected at a depth of 2 m, one mile off shore and 5 miles north of Milwaukee harbor. After 8 hrs, the radioactive water (225 ml) was put in one hundred 250 ml bottles. Five, 10 and 25 ml aliquots of each sediment extract were introduced to the bottles. Controls, i. e., radioactive water without sediment extract, were run with each harbor sediment. The bottles were kept in an environmental chamber under constant illumination at  $18.3 \text{ C}$ . Every 24 hrs a 50 ml sample was taken from each bottle and filtered through a 47 mm dia,  $0.45 \mu$  HA Millipore filter. The filter was rinsed with 5 ml of distilled water and glued on an aluminum planchet. After drying, the filters were placed in an atmosphere of concentrated HCl in a desiccator for 2 min to remove inorganically fixed carbon (Wetzel, 1965b). Then the planchets were placed in a Hamner thin-window gas-flow proportional counter and 2 one-min counts were made of the radioactivity.

#### E. Bioassay of Sediments Using *Daphnia pulex* and Zooplankton

##### 1. *Daphnia pulex*

The test sediments were diluted in the same way as in the phytoplankton tests, and 5, 10 and 25 ml amounts were added to dechlorinated city water in 125 ml bottles. A series of control tests, i. e., water without sediment, was also used. Approximately 12 adult *Daphnia pulex* (length  $> 400 \mu$ ) were placed in each bottle. The bottles were filled to the top and stopped with a small wad of moist cotton to prevent the crustaceans from becoming caught in the surface film. After 24 hrs some *Chlorella* was added as food. The experiment ran

for 48 hrs at a temperature of about 18 C. The number of live and dead adults (length  $> 400\mu$ ) and young (length  $< 200\mu$ ) were counted with the aid of a magnifying glass.

## 2. Zooplankton

The same procedure as for Daphnia pulex was used, except that filtered fresh water from Lake Michigan was used. Lake Michigan zooplankton was collected 4 miles east of Milwaukee harbor with a  $\frac{1}{2}$ -m net,  $350\mu$  mesh-size, towed at a depth of about 2 m at 2300 CDT. Lake temperature was 16.7 C and the tests were run at about 18 C. The experiment was terminated in 48 hrs. The contents of each bottle were filtered through a #153 Nitex net, and the living and dead Daphnia, other cladocerans, and copepods were counted. The number of Daphnia trapped in the surface film was also recorded. Daphnia consisted mainly of D. retrocurva with some D. longiremis, D. schodleri and D. galeata-mendotae. Bosmina sp. and Ceriodaphnia sp. were the most abundant other cladoceran genera present. Most of the copepods were Cyclops bicuspidatus and Diaptomus ashlandi with several other species of Diaptomus also present. Only Daphnia counts were used to determine the percentage of mortality, since they were most abundant and large enough in size for this determination.

### F. Bioassay of Sediments Using the Benthic Alga, Cladophora

Active growth of Cladophora could not be maintained in the laboratory until soil extract was added to the Gorham's culture medium. Soil extract was prepared by mixing garden soil 1 : 2 with distilled water, autoclaving the slurry at 15 lb pressure and 20 C for 20 min, and filtering the supernatant through No. 1 Whatman's filter paper.

### 1. Dilute, untreated sediment experiment

Sediments from stations B-3 and T-1 were diluted in the same way as for the phytoplankton bioassay tests and added to 50 ml of Gorham's medium in 60 ml bottles. Strands of Cladophora from Lake Michigan, which had been in ASM (poor in phosphates and nitrates) medium (McLachlan and Gorham, 1961), for 2 weeks, were placed in each bottle. Growth was to be determined by increase in wet weight, but sediment particles clung to the Cladophora and the experiment was ended after 48 hrs. The Cladophora was deteriorating. This method was considered unsatisfactory.

### 2. Sediment extract experiment

Sediment extracts were made as in the carbon-14 bioassay test. Two 100 × 15 mm glass petri dishes were set up for each harbor sediment, the Rouge River hopper dredge sample, and Fuller's Earth. Fifty ml of Gorham's medium fortified with the soil extract was put in each dish. Ten ml and 25 ml of each sediment extract were added. Ten dishes were left without sediment extract as controls. Short strands of Cladophora, which had been in ASM medium for 2 weeks, were cut, measured, and placed in each petri dish. Strands with little or no branching were selected. The petri dishes were kept in an environmental chamber under constant illumination at a temperature of 18.3 C for 7 days. At termination of the test, the Cladophora strands were remeasured under a low power microscope.

## IV. RESULTS AND DISCUSSION

### A. Selection of Sediments by Benthic Organisms

These studies are useful in determining the possible effects of open-lake disposal on benthic organisms at the dumping site. The organisms may assume random distribution in the different sediment types, or selection of certain sediments in preference to others may occur. The distribution of Pontoporeia in the test sediments showed a definite preference for the open-lake (O) sediments during the first trial (Fig. 11). Sixty-two per cent (58 organisms) of the Pontoporeia were recovered in this sediment. The next highest concentration was 6 organisms in the outer harbor sample from Calumet (C-1). Four were recovered from the T-5 sample and 3 from the GB-4. Only 1 or 2 Pontoporeia were found in the other 13 samples that contained Pontoporeia. Fifty-eight per cent of the sediments did not have any Pontoporeia.

The Gammarus, which were introduced with Pontoporeia, were not as selective of sediments. The greatest number in one sample was 4. Four were found in both the open-lake (O) and Great Sodus Bay (S-2) sediments. Three were in the Calumet (C-1) sample. One or 2 Gammarus were recovered from 11 other samples.

In the second trial, only Pontoporeia were used. Of the organisms recovered, 31.9 per cent were in the open-lake sample and 35.5% in the Great Sodus Bay (S-2) sample (Fig. 12). The Calumet (C-1) sediment had 9.2%. Fifty per cent of all the samples did not have any Pontoporeia.

Despite limited mobility, the midge larvae also showed a preference for some sediments and avoided others in the first trial (Fig. 13). The greatest number of larvae recovered from a single sample was 19 from Calumet (C-1). The open-lake (O), Toledo (T1, T-3) and

C-1 6-0	CL-4 0-0	I-4 0-0	T-5 4-0	B-5 1-0
CL-1 0-0	S-1 1-0	GB-3 0-0	C-3 0-0	GB-1 0-0
GB-5 0-0	R-1 0-0	T-3 0-0	B-4 0-0	Control 2-0
T-1 2-0	C-2 0-0	CL-5 1-0	I-3 0-0	M 0-0
R-4 0-0	GB-2 1-0	CL-2 0-0	R-2 0-2	B-1 0-0
I-2 0-1	T-2 0-0	C-4 0-0	B-3 1-0	S-2 2-0
R-3 0-1	B-2 0-0	CL-3 0-0	C-5 2-1	I-5 0-0
I-1 2-1	GB-4 3-1	T-4 0-0	O 51-7	R-5 0-0

Fig. 11. Distribution of amphipods in sediments (Pontoporeia and Gammarus), first trial. [ Letter and number indicate harbor and sediment. Lower numbers in each box indicate live and dead organisms. ]

C-1 12	CL-4 0	I-4 0	T-5 0	B-5 1
CL-1 0	S-1 2	GB-3 5	C-3 0	GB-1 1
GB-3 0	R-1 0	T-3 1	B-4 6	Control 2
T-1 0	C-2 0	CL-5 2	I-3 0	M 0
R-4 0	GB-3 3	CL-2 0	R-2 1	B-1 0
I-2 0	T-2 0	C-4 2	B-3 1	S-2 39
R-3 0	B-2 0	CL-3 1	C-5 3	I-5 1
I-1 2	GB-4 1	T-4 0	O 44	R-5 0

Fig. 12. Distribution of amphipods (Pontoporeia) in sediments, second trial. [ Letter and number indicate harbor and sediment. Lower number in each box indicates number of live organisms. No organisms were dead. ]

Great Sodus Bay (S-1 and S-2) sediments all had 10 or more larvae. All of the Green Bay samples contained midges. Twenty-three sediments did not have any larvae. The occurrence of midges in a sediment did not indicate that it was a suitable environment for them, since the mortality was 100 per cent in 5 samples.

In the second trial, 24 per cent of the midges were recovered from the Green Bay samples and 15.6 % from the Toledo samples (Fig. 14). The Calumet sediments each had at least 12 larvae, except for C-3. The open-lake and Sodus Bay (S-1, S-2) samples had 22, 18, and 13 larvae, respectively. The Rouge River and Indiana Harbor sediments contained the fewest midges. The highest mortality (85.7%) was in the Rouge River hopper dredge sample.

It is apparent that the organisms were selective of sediments. Selection was not a function of experimental design, i. e., the organisms were not concentrated in one part of the aquarium. They were concentrated in certain sediments, which can be assumed to most closely satisfy the environmental requirements of the organisms or at least did not contain substances detrimental to their existence. The Pontoporeia selected the cleanest sediments that contained some sand. The midge larvae, Chironomus tentans, are more pollution tolerant than Pontoporeia and they are usually associated with organic sediments as indicated by their occurrence in the sediments of some of the harbors. Midge larvae are not found in environments that are badly polluted, however. Certain sediments were avoided by or resulted in high mortality of the test animals and were obviously not suitable environments for either the amphipods or the more pollution tolerant midge larvae. These sediments were from Buffalo (B-2), Calumet (C-3), Cleveland (CL-2), Indiana Harbor (I-3, I-4), Milwaukee (M), Rouge River (R-1, R-4), and Toledo (T-2, T-4).

C-1 19-0	CL-4 2-0	I-4 0-0	T-5 5-0	B-5 5-0
CL-1 1-0	S-1 10-2	GB-3 2-0	C-3 0-0	GB-1 0-0
GB-5 5-2	R-1 0-1	T-3 11-0	B-4 1-2	RH 0-0
T-1 13-0	C-2 0-5	CL-5 3-0	I-3 0-5	M 1-0
R-4 0-0	GB-2 9-2	CL-2 1-0	R-2 0-0	B-1 1-0
I-2 0-1	T-2 0-0	C-4 2-2	B-3 4-2	S-2 14-0
R-3 0-0	B-2 0-2	CL-3 0-0	C-5 0-0	I-5 3-3
I-1 1-0	GB-4 6-0	T-4 0-0	O 16-0	R-5 5-0

Fig. 13. Distribution of midge larvae, Chironomus tentans, in sediments, first trial. [ Letter and number indicate harbor and sediment. Lower numbers in each box indicate live and dead midges. ]

CL-5 6-0	O 22-0	B-3 1-1	B-2 4-0	B-1 6-0
GB-1 14-2	T-3 12-2	C-4 14-3	CL-2 1-1	R-3 0-0
C-5 11-5	S-1 14-4	T-5 25-0	S-2 13-0	M 6-0
T-2 22-2	RH 1-6	GB-2 15-2	R-2 0-0	B-5 4-0
C-1 17-0	CL-1 7-3	GB-4 22-3	F 8-0	I-4 0-0
CL-3 1-0	I-5 1-0	C-3 2-1	R-5 6-1	T-4 4-0
I-3 1-2	Control 8-0	GB-5 13-2	T-1 10-1	B-4 3-1
R-4 6-0	I-2 1-1	C-2 12-0	CL-4 8-0	R-1 3-1
		GB-3 23-0	I-1 5-5	

Fig. 14. Distribution of midge larvae, *Chironomus tentans*, in sediments, second trial. [ Letter and number indicate harbor and sediment; lower numbers in each box indicate live and dead midges. ]

## B. Benthos Viability Studies

These studies are pertinent to understanding what the direct effect of dredgings disposed of in the lakes may have on the biota, since the organisms used in the tests are bottom dwellers. Various midge larvae and the amphipod, Gammarus lacustris, would represent the biota in many of the shallow areas presently used for disposal. Pontoporeia affinis occurs in the deeper, colder waters, but results of their response to sediments should be useful for understanding the consequences of disposal in deep areas. Furthermore, P. affinis is considered to be a clean water organisms and intolerant of pollution, whereas many midge larvae are pollution tolerant.

### 1. Amphipods

No Pontoporeia died in the control sediments in the 24 hr test, but one dead each was found in the open lake and Fuller's Earth controls in the 48 hr test (Fig. 15). Every harbor had some sediments with a mortality significantly greater than the controls. The lowest mortality occurred in the Great Sodus, Green Bay, and Toledo harbor sediments. The greatest mortality was in the Rouge River and Indiana harbor sediments. The sample from the hopper dredge (Rouge River) resulted in 100% mortality in both the 24 and 48 hr tests. In general, sediments from the rivers were more toxic than those from the outer parts of the badly polluted harbors.

An insufficient number of Gammarus was used in the tests to develop a valid comparison of their response and that of Pontoporeia to the sediments. Only 33 Gammarus were used as compared with 128 Pontoporeia. The total mortality was 9% for Gammarus, which suggests that this organism may be more tolerant of the test conditions than were Pontoporeia, which exhibited 70% total mortality.

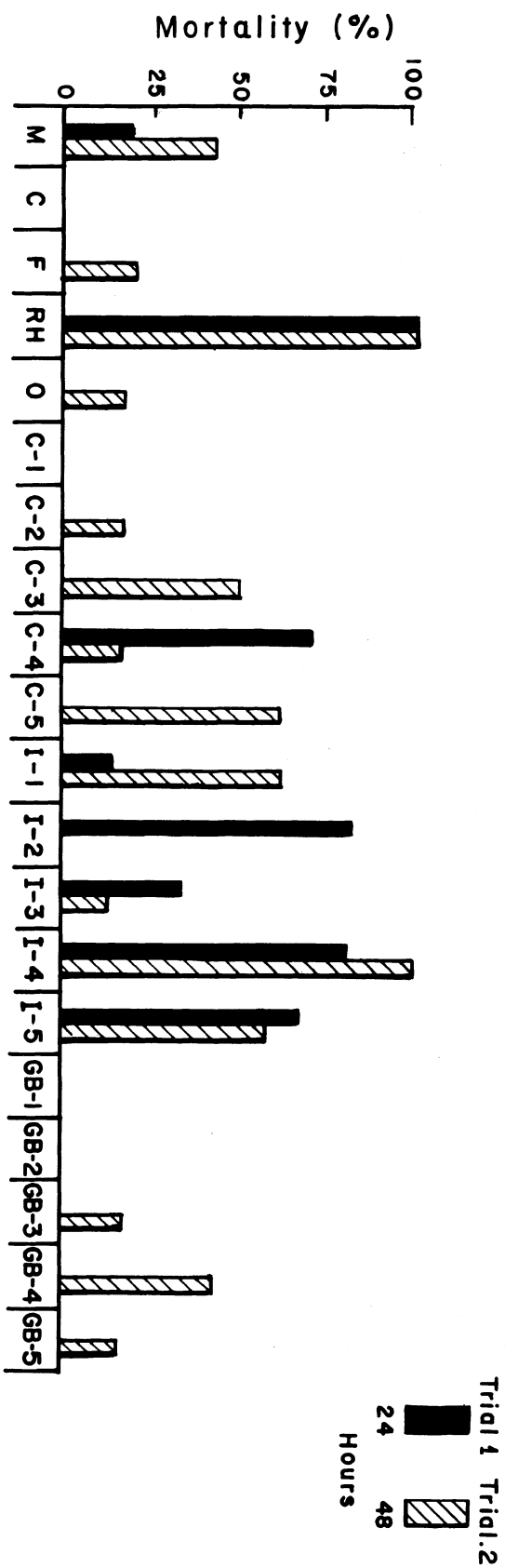
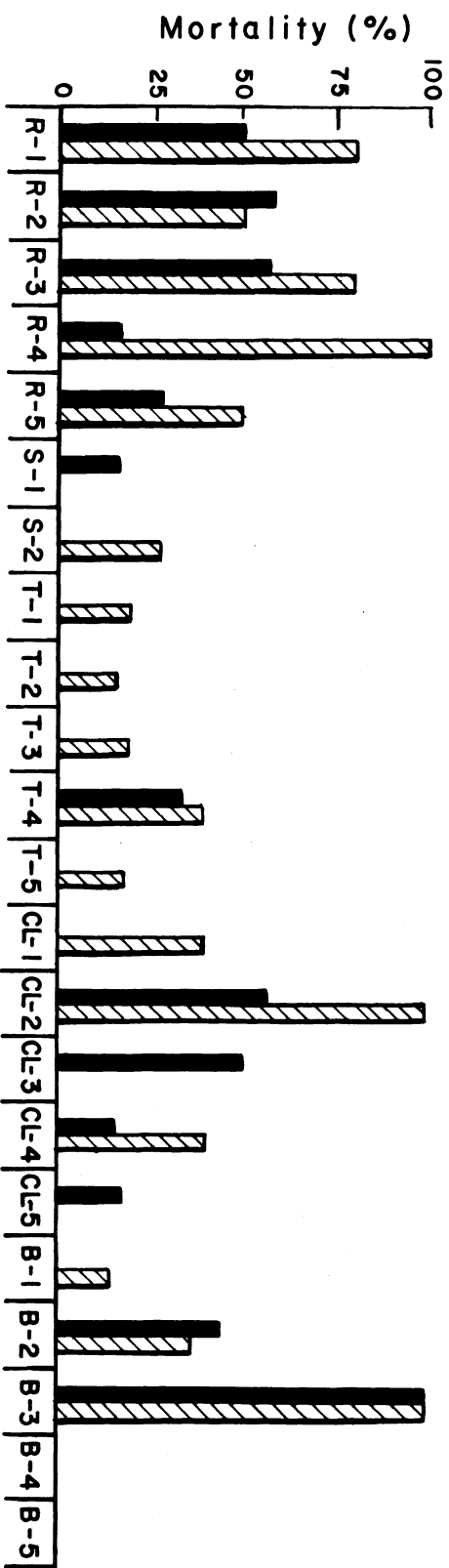


Fig. 15. Percentage mortality of amphipods in various harbor sediments. *Pontoporeia affinis* was used in the 24 hr tests, and both *P. affinis* and *Gammarus lacustris* were used in the 48 hr test. Numbers refer to sampling locations in harbors, which are designated by letter.

### Buffalo

The outer harbor sediments did not cause significant mortality. River sediments were toxic, especially from station B-3 with 100% mortality in both the 24 and 48 hr tests.

### Calumet

The outer harbor sediments did not result in any significant mortality in either the 24 or 48 hr tests, since the 16.7% mortality in C-2 sediments in 48 hrs was not higher than the mortality in the controls. Results of the tests are not in agreement for the river sediments, since Pontoporeia died only in the C-4 sediments in the 24 hr tests. They died in all three of the river sediments in the 48 hr test, although the mortality in the C-4 sample was not significant.

### Cleveland

Differences in mortality in sediments from the outer harbor and river were not well established. The outer harbor sediment from CL-5 was not toxic, but 42.8% of the Pontoporeia died in 48 hrs in the CL-4 sample. The sample from station CL-2 in the Cuyahoga River resulted in 100% mortality in 48 hrs. A 40% mortality occurred in 48 hrs in the river sediment from CL-1. The mortality was 50% in 24 hrs in the CL-3 sample, but no mortality occurred in the 48 hr test.

### Great Sodus Bay

Mortality in the sediments from the two stations in the bay was not significantly greater than the control tests, although the mortality was 28.5% in 48 hrs at S-2.

### Green Bay

Mortality was very low in sediments from all the stations. The only significant mortality (42.8%) occurred in the sample from station GB-4 one mile north of the river mouth.

### Indiana Harbor

Mortality was significant in all the sediments in the 24 hr test. The 48 hr test did not substantiate the high mortality of 83.3% which occurred in the I-2 sample in 24 hrs. The highest mortality was in the sediments from the innermost part of the harbor (stations I-4 and I-5). The lowest mortality was in the sediment from the largest part of the harbor (I-3).

### Rouge River

All the sediments were toxic to Pontoporeia and resulted in mortalities of 50% or more in 48 hrs. Mortality was less than 30% in samples R-4 and R-5 in 24 hrs, which nevertheless was significant, especially in view of the 48 hr test results.

### Toledo

Mortality was not significant in any sediment, except in sample T-4, which was collected in the vicinity of the Toledo municipal sewage treatment plant.

## 2. Midge larvae

The midge larvae did not die in either the clean sand or open-lake sediment (Fig. 16). Mortalities of 33.3% and 16.7% occurred in the Fuller's Earth controls in, respectively, the 24 and 48 hr tests. All the harbors had sediments which caused mortality significantly greater than the controls. The overall results were closely similar to those of the amphipod experiments, although the percentage of midges that died was much higher than the per cent mortality of amphipods. The higher mortality of the midge larvae may be due to their more limited ability to move or it may be because of the smaller containers used in the midge experiment. The smaller containers probably restricted the flow of water in and out; oxygen depletion may have occurred. The dead larvae

were usually found on the surface of the mud, and there were often covered with an oil film.

#### Buffalo

Mortality was high in both the river and outer harbor sediments. It was 50% or more in all sediments in 24 hrs and 100% in sediments from B-1, B-3, and B-4 in 48 hrs.

#### Calumet

Mortality was 50% or more (100% at C-2) in all sediments in 24 hrs. None of the midges died in the C-1 sample, and mortality was less than 50% in samples C-4 and C-5 in the 48 hr test.

#### Cleveland

The river sediments resulted in a mortality of 50% or more, and the mortality in outer harbor samples was 25% or less in 24 hrs. The percentage mortality was 50% or greater in all sediments in 48 hrs, except for the CL-2 sample where no dead larvae were present.

#### Great Sodus Bay

The results indicate substantial difference between the sediments collected in the Coal Dock Channel (S-1) and in the harbor entrance (S-2). None of the midges died in the S-2 sediment, but the mortality was 50% in 24 hrs and 100% in 48 hrs in the S-1 sample.

#### Green Bay

Mortality was not significant in any of the sediments, except in the sample from GB-5 near the bay spoil area. The percentage mortality was 33.3 in 24 hrs and 83.3 in 48 hrs. This greater mortality at station GB-5 may be due to dredging spoil washing out of the nearby diked disposal area, since the dikes washed out in several places.

### Indiana Harbor

Mortality was greater in the sediments from this harbor than in any of the other harbors. It was greater than 60% for all samples in both the 24 and 48 hr tests.

### Rouge River

More than 60% of the midges died in 24 hrs, except in sample R-5 with a 40% mortality. Mortality was 100% in sediments from R-2, R-3, and R-4 in 48 hrs and mortality at R-5 was 80% in 48 hrs.

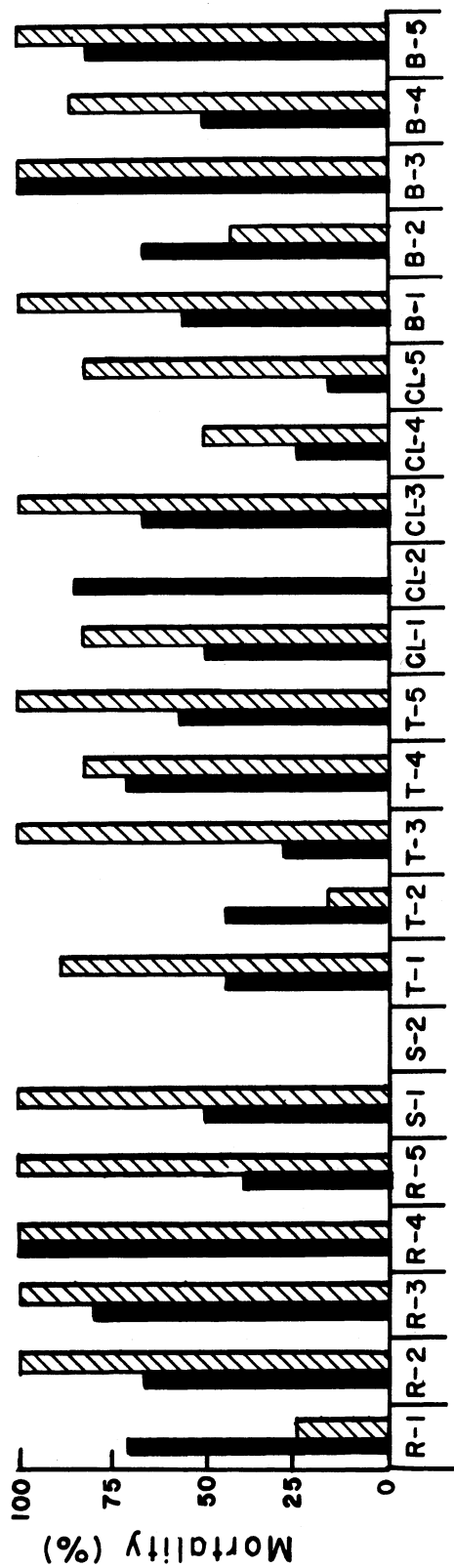
### Toledo

The greatest mortality in 24 hrs was in the T-4 sample (71.4%). The percentage of dead midges was greater than 80% in all sediments, except T-2, in 48 hrs.

### C. Bioassay of Sediments Using Phytoplankton and Algal Cultures

These experiments were designed to determine the effect of harbor sediments on the growth of phytoplankton, and the laboratory procedure was such as to simulate the possible effects of open lake disposal of dredgings on the plankton. The use of a photometer to measure changes in the number of plankton cells had definite disadvantages in that different operators did not get the same optical density readings. Some great fluctuations in the data can be attributed to this problem. It would not appreciably affect the significant trend that developed over the entire period of the tests, however.

The predominant trend that is apparent is a decrease in the optical density during the period of the experiments. In most tests, the greater the concentration of sediment, which had been added initially to the test flasks, the greater the decrease in optical density. This result can be



Trial 1 Trial 2  
 24 48  
 Hours

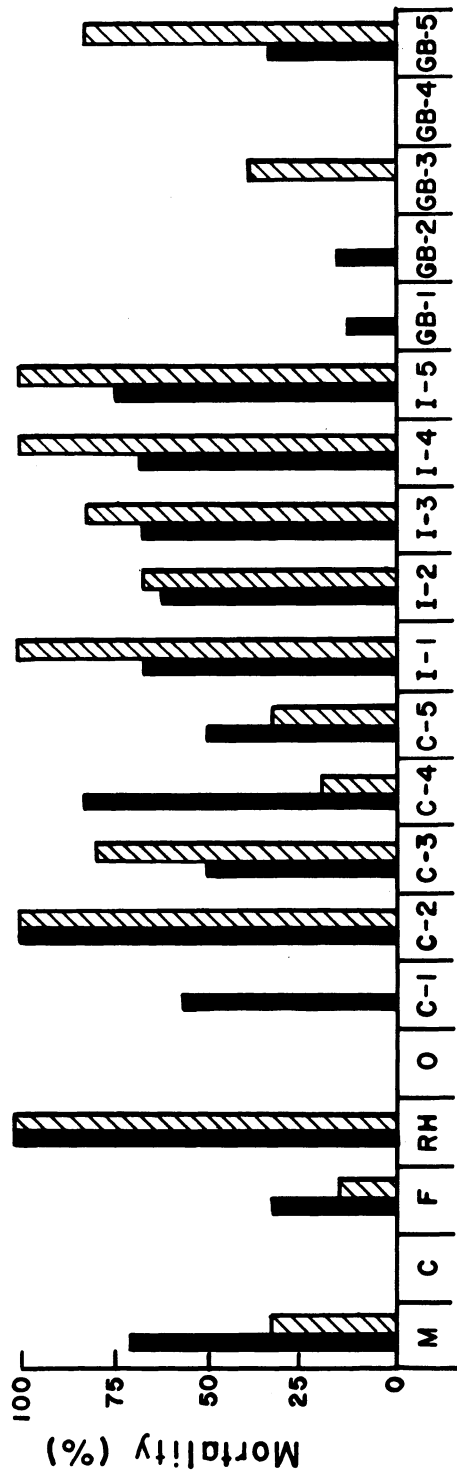


Fig. 16. Percentage mortality of midge larvae (*Chironomus tentans*), in various harbor sediments. Numbers refer to sampling locations in harbors, which are designated by letter.

interpreted as indicating a decrease in the number of individual cells, but not necessarily a decrease in growth. The decrease in optical density could be due to some clumping of sediment particles and algal cells. It if actually represents a decrease in growth, then this probably would be due to the decrease in amount of available light because of the turbidity caused by the sediment. Nevertheless, the results are not conclusive, other than to suggest that some reduction, probably temporary, in the numbers of algal cells may occur in dredging and open-lake disposal areas.

#### 1. Phytoplankton from Juneau Park Lagoon, Milwaukee Wis.

Twenty-nine control tests were made. The optical density of 5 did not change, 5 had a slight increase, 13 showed a slight decrease, and only 4 had any major increase or decrease (U.S. Army Corps of Engineers, 1969). Almost all of the tests with sediments followed the same general trend of the greater the concentration of sediments the greater the decrease in optical density. The data for the experiment with the Calumet sediments are presented as examples (Figs. 17-19). It should also be noted that the results of the tests with Fuller's Earth were similar to the Calumet results (Fig. 17).

#### 2. Algal Cultures

Anabaena and Chlorella were used in these experiments.

Controls, i. e., without sediment, were included with the tests of each set of harbor sediments. In general, the optical densities of the controls remained essentially the same throughout the period of the experiment, indicating no appreciable change in numbers of algal cells.

Only the results of the tests for Calumet, Milwaukee and Great Sodus Bay, as well as the hopper dredge and Fuller's Earth samples, are presented graphically (Figs. 20 - 21).

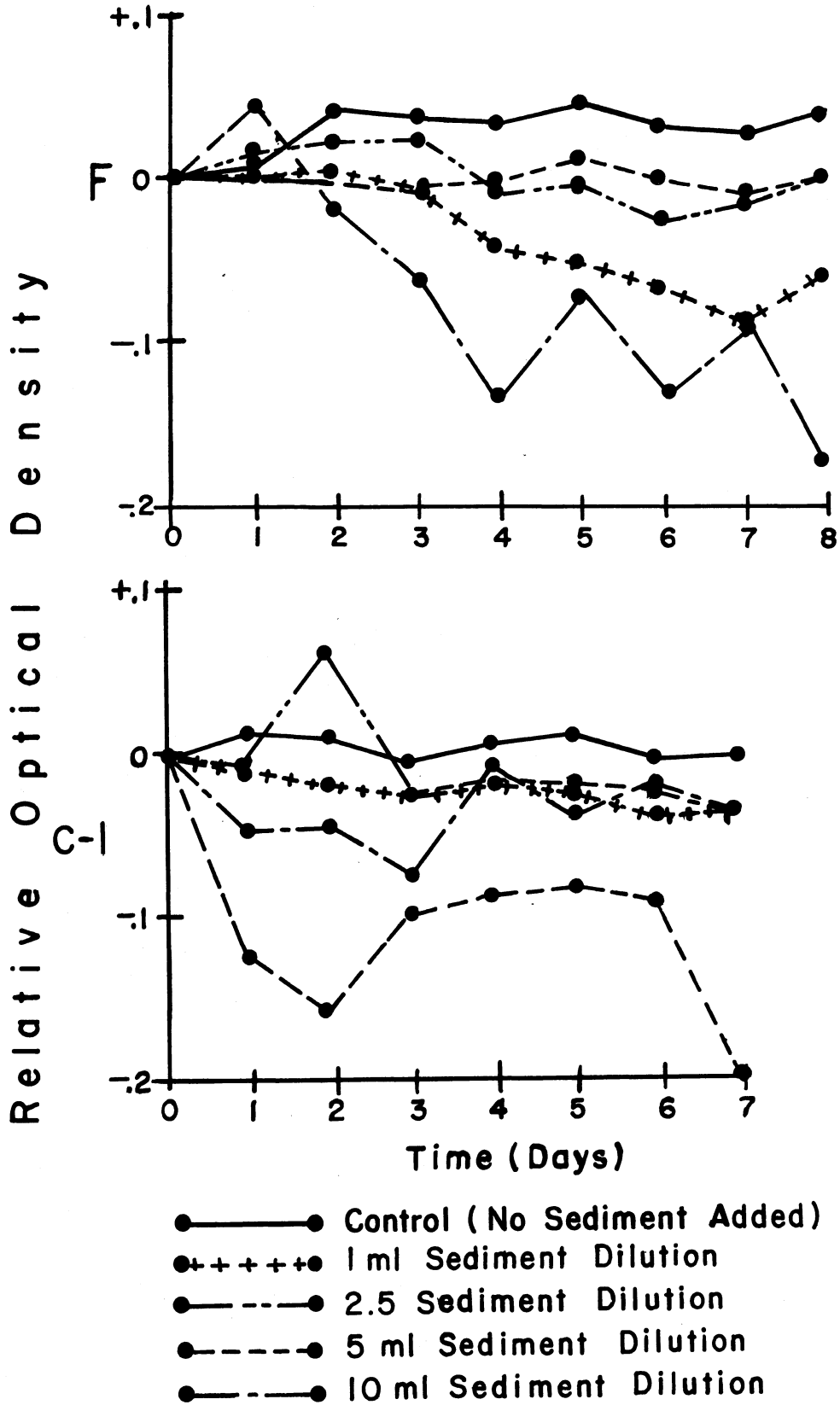


Fig. 17. Response of phytoplankton from Junean Park Lagoon to various concentrations of sediments (1-10 ml) from Calumet Harbor, Ill., as measured by change in optical density. (Letter "C" indicates Calumet and the numbers denote the sampling stations; "F" indicates the Fuller's Earth control sample.)

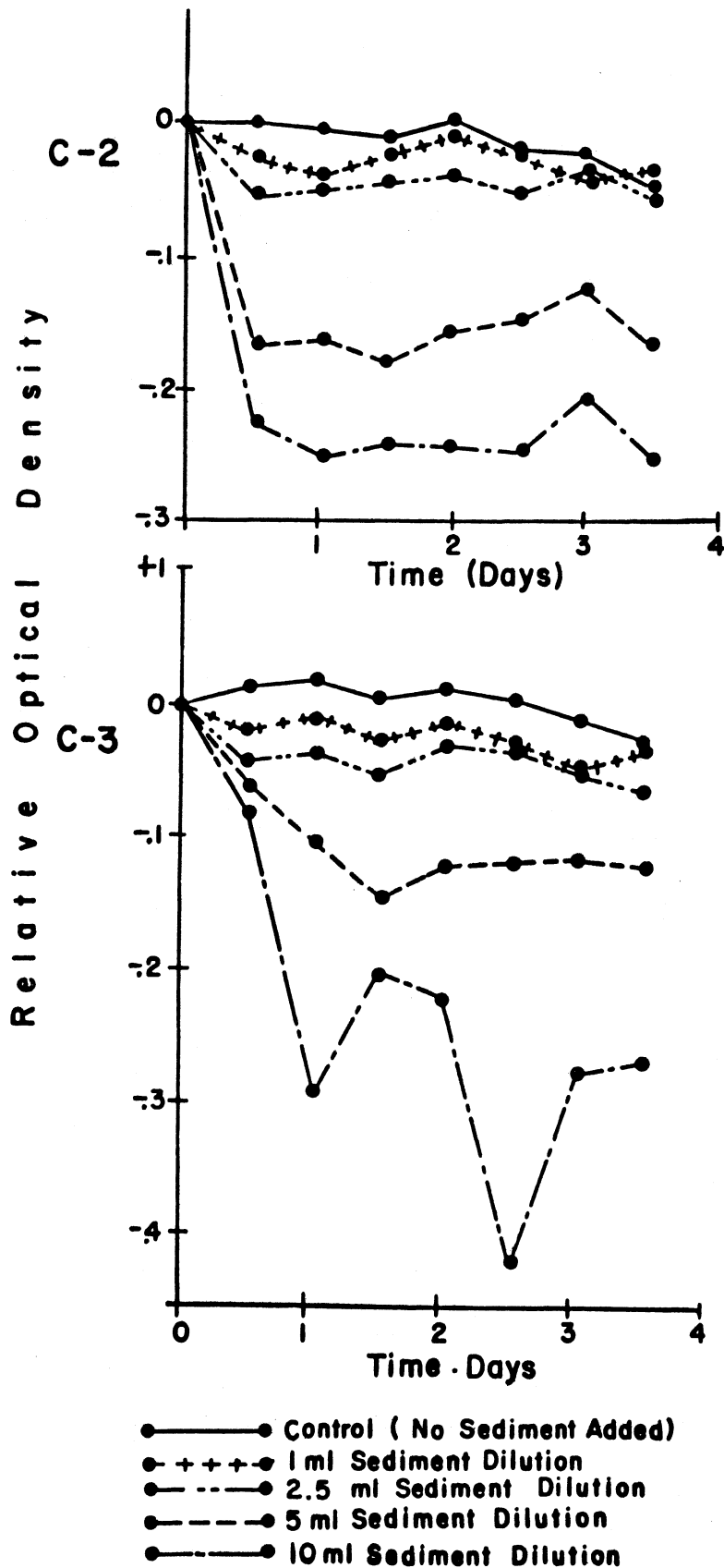


Fig. 18. Response of phytoplankton from Juneau Park Lagoon to various concentrations of sediments (1-10 ml) from Calumet Harbor, Ill., as measured by change in optical density. (Letters 'C' indicate Calumet and the numbers denote the sampling stations.)

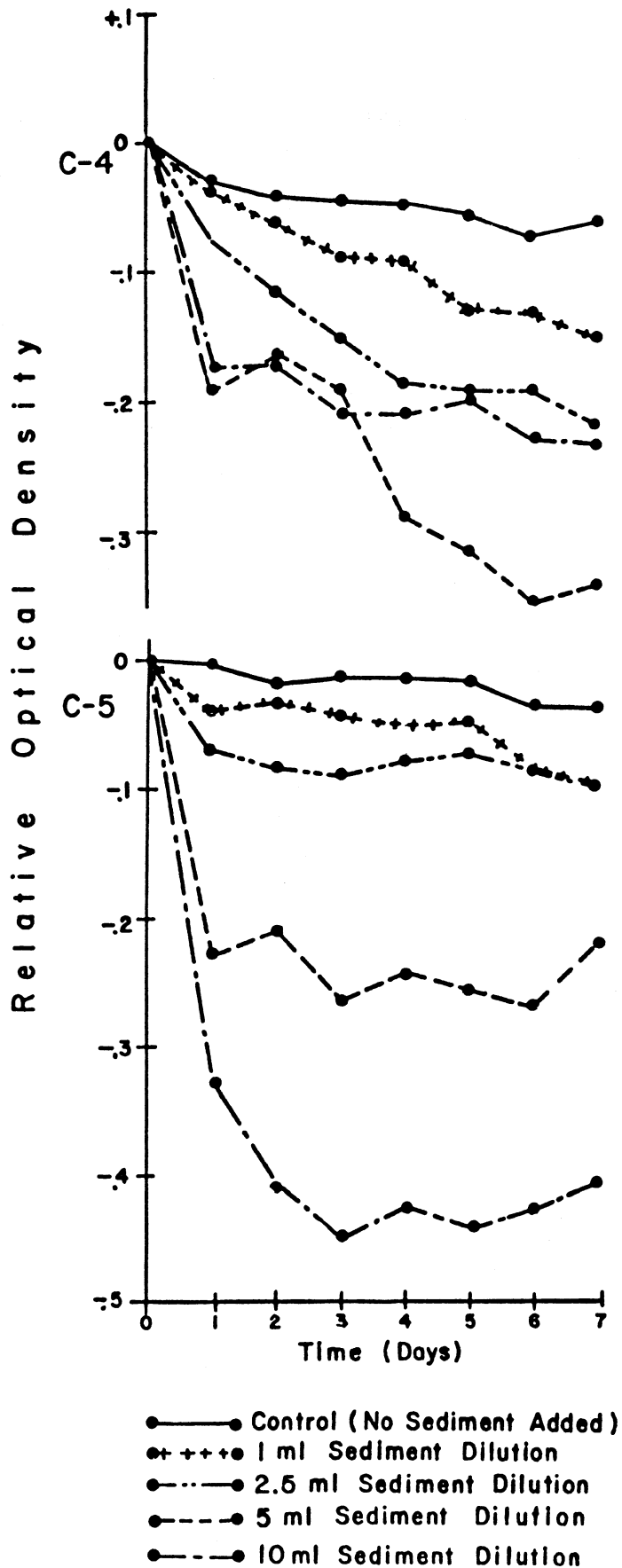


Fig. 19. Response of phytoplankton from Juneau Park Lagoon to various concentrations of sediments (1-10 ml) from Calumet Harbor, Ill., as measured by change in optical density. (Letters "C" indicate Calumet and the numbers denote the sampling station.)

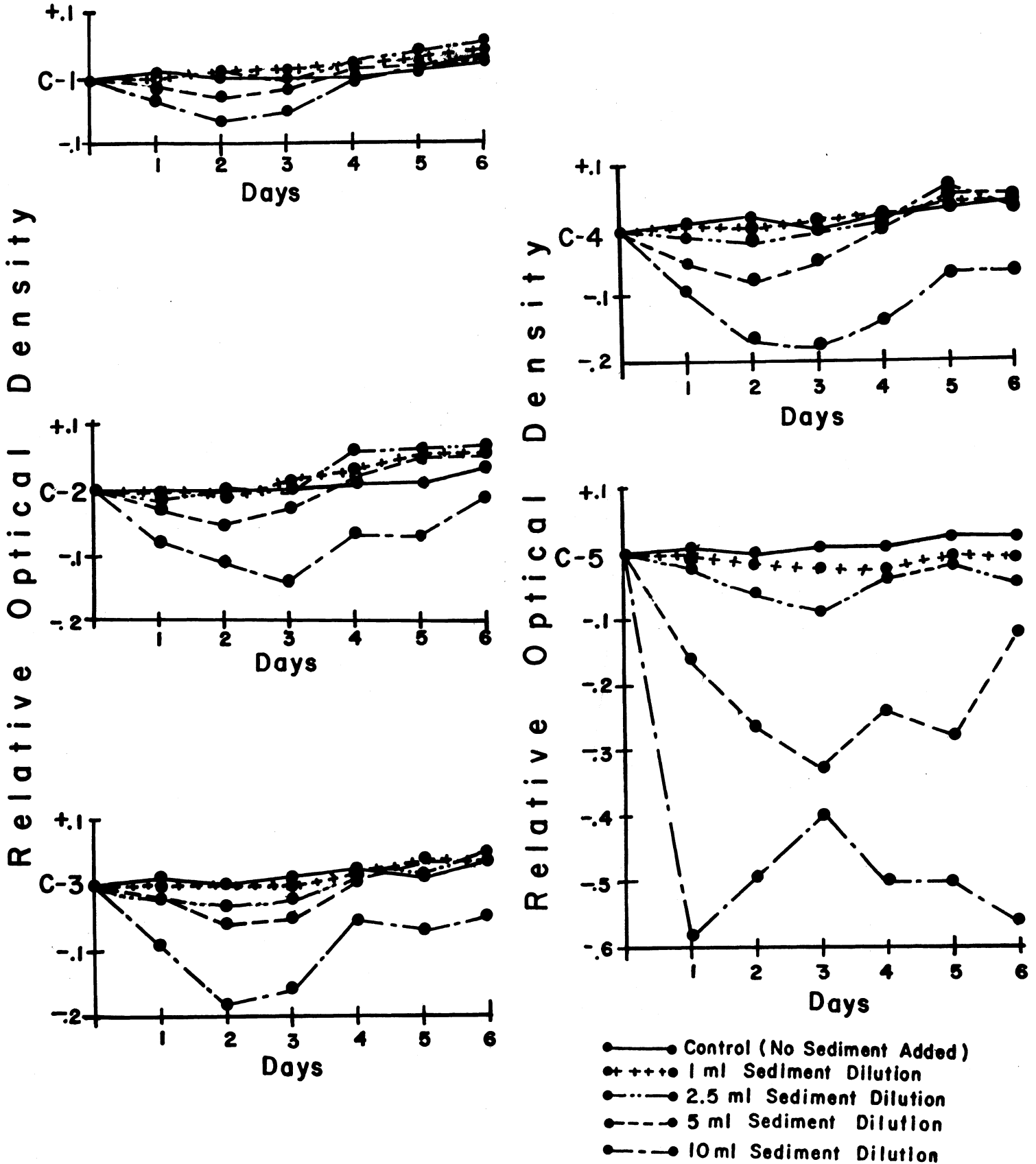


Fig. 20. Response of cultures of *Anabaena flos-aquae* to various concentrations (1-10 ml) of sediments from Calumet harbor, Ill., as measured by change in optical density. (Letter "C" indicates Calumet, and numbers indicate station.)

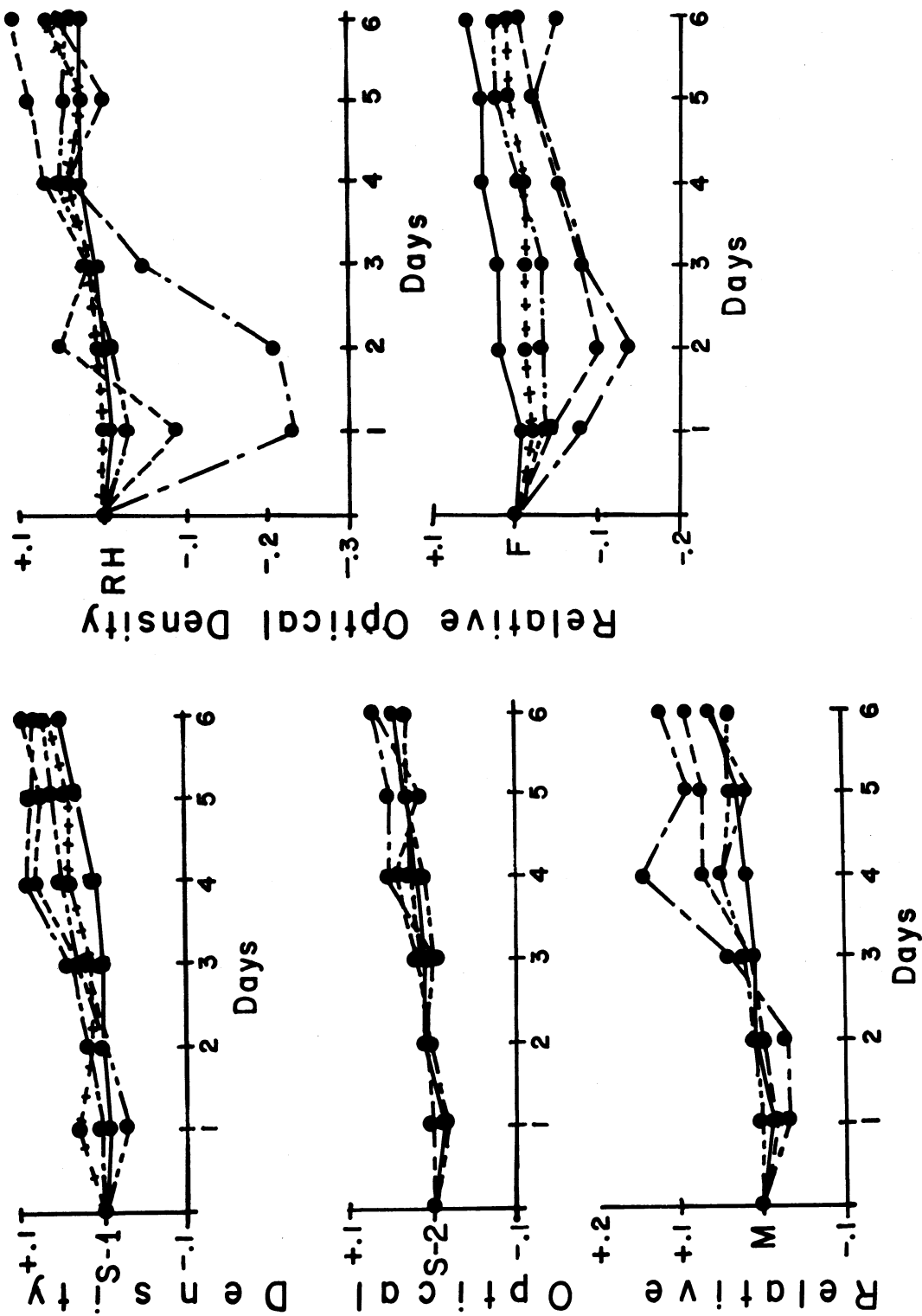


Fig. 21. Response of cultures of *Anabaena flos-aquae* to various concentrations (1-10 ml) of sediments from various harbors, as measured by change in optical density. (Letter "S" denotes Great Sodus Bay harbor, N.Y.; "M" - Milwaukee harbor, Wis.; "RH" - Rouge River hopper dredge sample; and "F" - Fuller's Earth sample.

The general response of the Anabaena cultures, which had sediments added to them, was for a decrease in optical density and then a subsequent increase. This response was significant in the test with the higher concentrations of sediments (5 and 10 ml). The cultures with the lower concentrations of sediment (1 and 2.5 ml) usually behaved like the controls, or optical density increased only slightly (Fig. 20). This general response occurred in almost all of the tests, except for Great Sodus Bay (Fig. 21) sediments. The tests with the Fuller's Earth (Fig. 21) behaved similarly to those with harbor sediments, so it is likely that the changes are not due to nutrients being available in the sediments, and the changes in the cultures may be related more to physical conditions, such as decrease in available light or clumping of sediment particles and algal cells.

Most of the Chlorella cultures, which had sediments added to them, had a decrease in optical density—the greater the concentration of sediment the greater the decrease in optical density, as shown by the results of the Calumet tests (Figs. 22-24). The test flasks with sediment from S-2 (Fig. 24) had an increase in optical density. The different response of the Chlorella to the S-2 sediment suggests some factor other than merely the physical as being important. The decrease in the optical in the majority of the tests may be due to the clumping of the highly organic sediment particles and the algae. Sediment from S-2 contained appreciable sand as well as organic material, and the increase in optical density may represent actual growth and lack of clumping. On the contrary, the test using station S-1 sediment had a decrease in optical density. This station is located by a coal dock, and possibly substances toxic to algal growth were in the sediment.

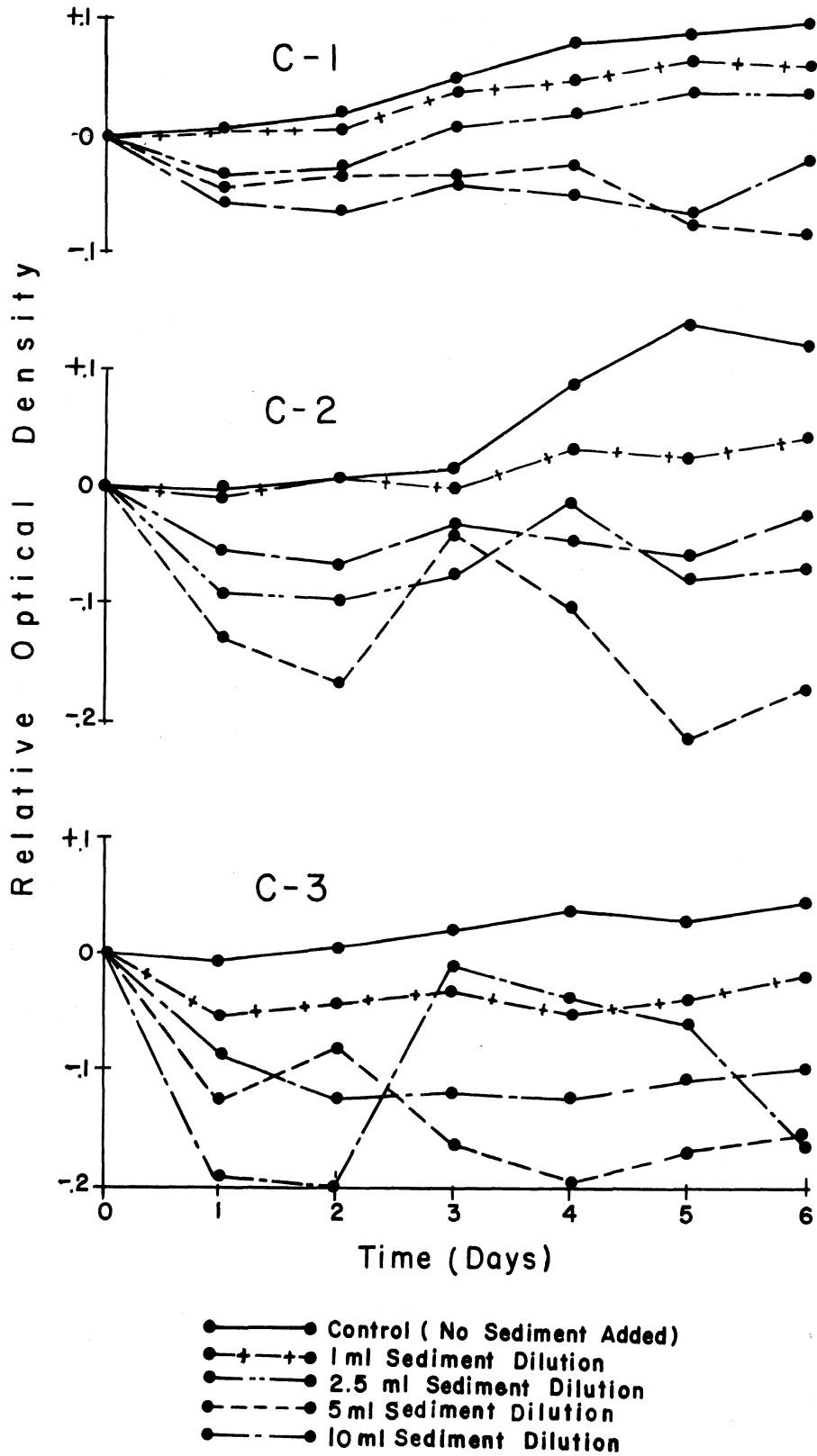


Fig. 22. Response of cultures of *Chlorella pyrenoidosa* to various concentrations (1-10 ml) of sediments from Calumet Harbor, Ill., as measured by change in optical density. (Letter "C" indicates Calumet, and numbers indicate station.)

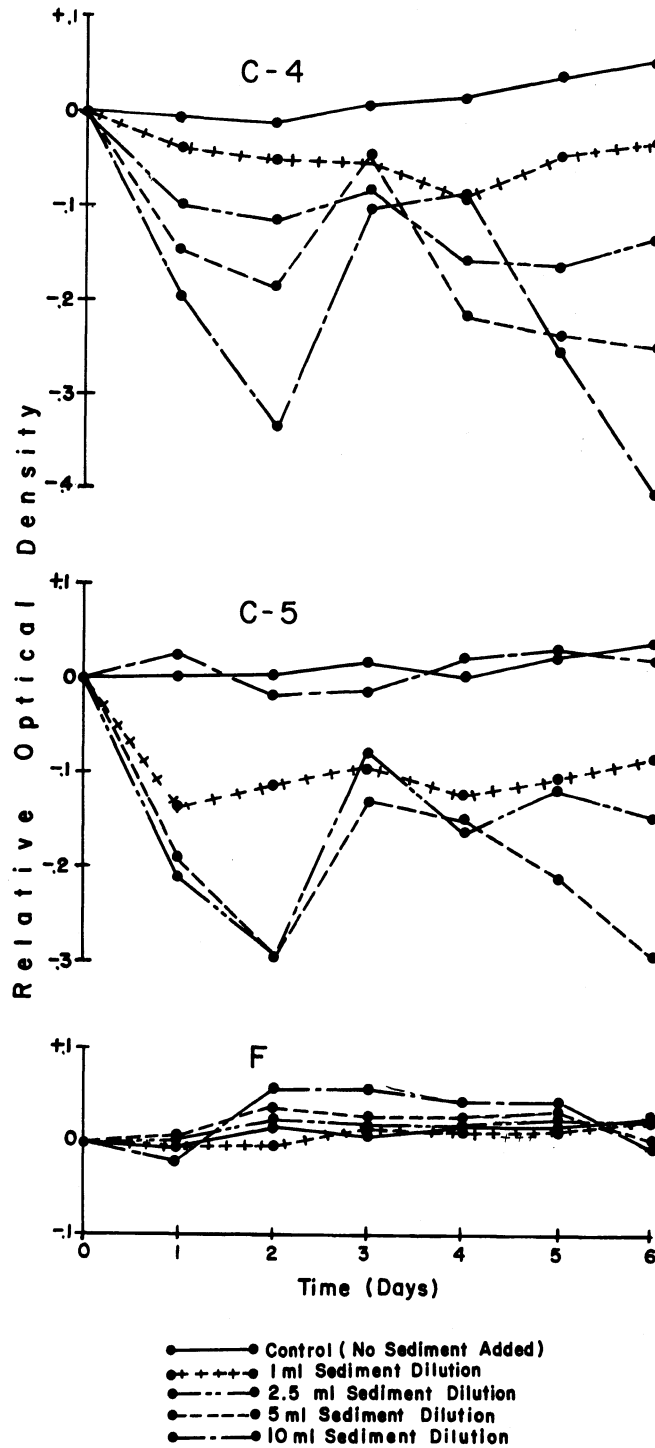


Fig. 23. Response of cultures of *Chlorella pyrenoidosa* to various concentrations (1-10 ml) of sediments, as measured by change in optical density. (Letter "C" indicates Calumet harbor, Ill., and "F" indicates Fuller's Earth; numbers denote stations.)

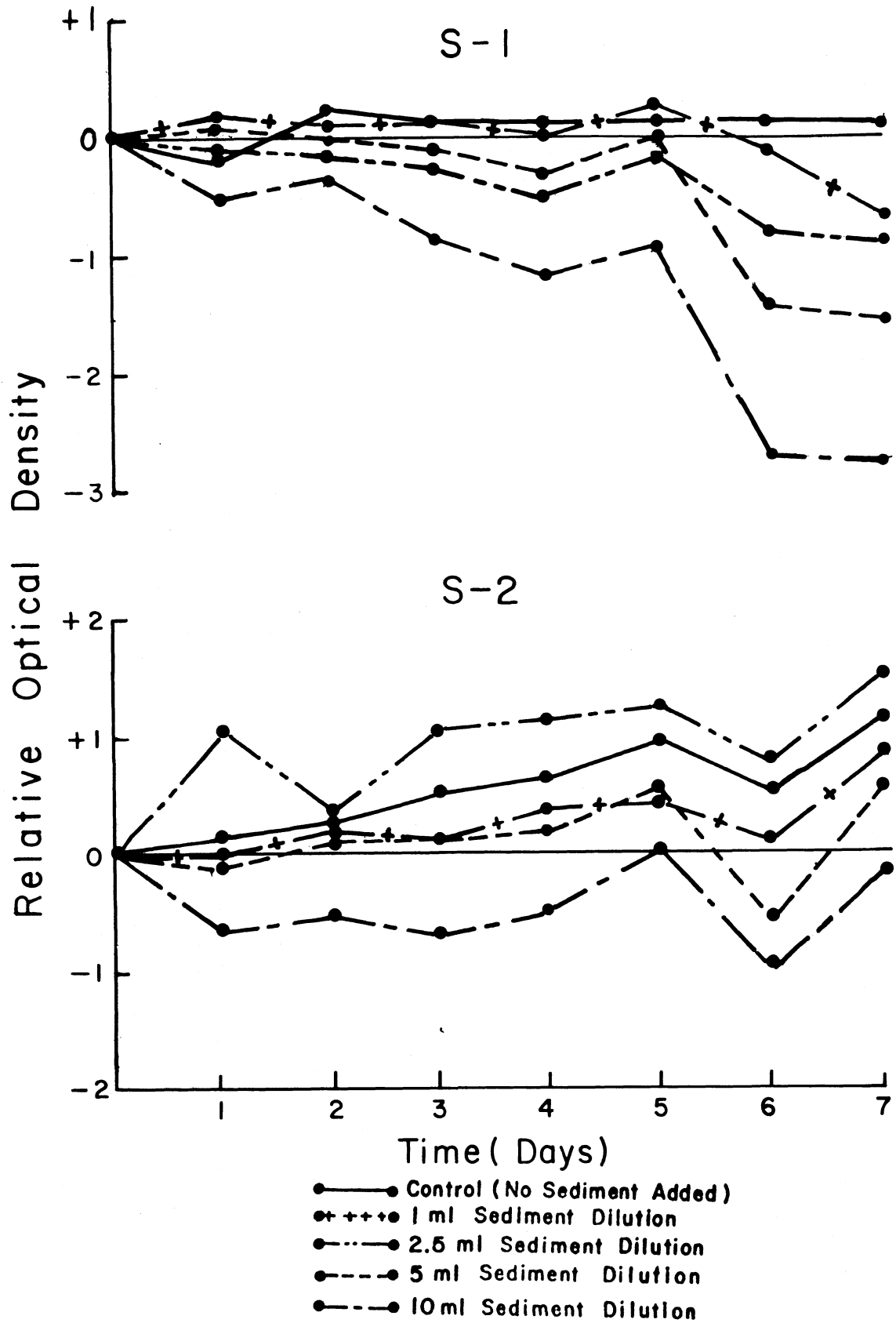


Fig. 24. Response of cultures of *Chlorella pyrenoidosa* to various concentrations (1-10 ml) of sediments, as measured by change in optical density. (Letter "S" indicates Great Sodus Bay harbor, N.y., and numbers denote stations.)

#### D. Bioassay of Sediments Using Carbon-14 Uptake by Phytoplankton

The purpose of this experiment was to determine the influence of harbor sediments on the growth of Lake Michigan phytoplankton. Carbon-14 has been successfully used to determine the productivity of aquatic systems, and its use is well established and accepted as a valid method (Strickland, 1960). Its use in the bioassays proved to be far more satisfactory than the measurement of change in cell numbers by change in optical density. Extracts of the sediments were used to avoid the possible clumping of algal cells and sediment particles, and adsorption of carbon-14 on sediment particles. The results of the carbon-14 tests indicate what may be happening under natural conditions when overflow water from the dredges enters the harbor and lake or when various components of the dredgings go into solution in the disposal area.

The carbon-14 method used in these bioassays demonstrated that extracts of many of the harbor sediments did stimulate the productivity of phytoplankton. The results are especially significant because natural Great Lakes phytoplankton was used. All the sediments were not included in this experiment, since the method and procedure was not developed in time to allow inclusion of all sediments. Nevertheless, enough samples were included from each harbor to be representative (Figs. 25-33). Fuller's Earth was used as a control and did not increase the uptake of carbon-14 (Fig. 25).

The uptake of carbon-14 was high in relation to the controls for fourteen of the sediments (all of the Buffalo, Calumet and Toledo, and GB-1, R-4, R-5, and S-1). Little or no uptake of carbon-14 occurred in the tests with nine sediments (all of the Cleveland and Indiana, and GB-5, R-1, RH, and S-2). No definite relationship could be established between carbon-14 uptake and concentrations of nutrients (Table 3). The tests with the highest

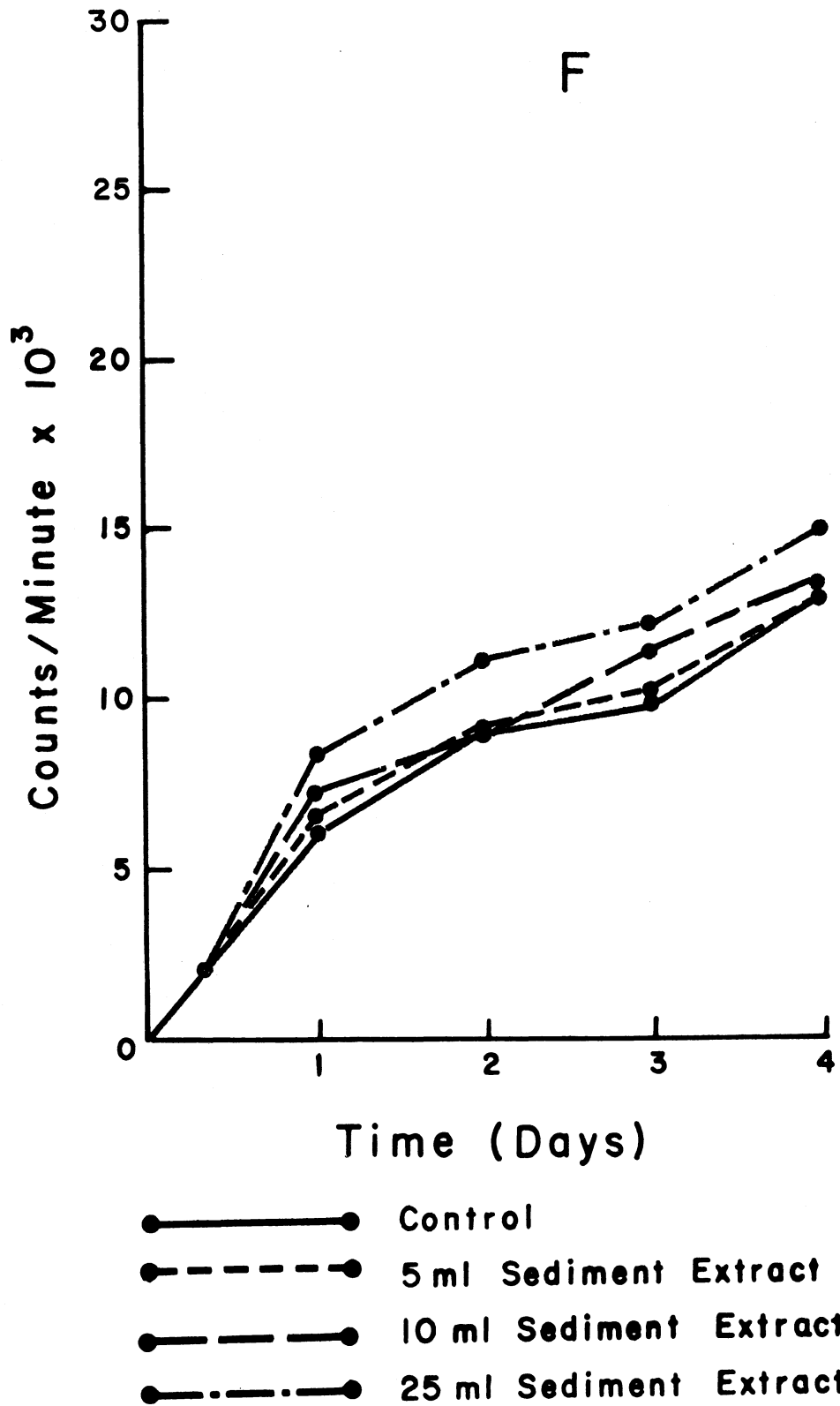
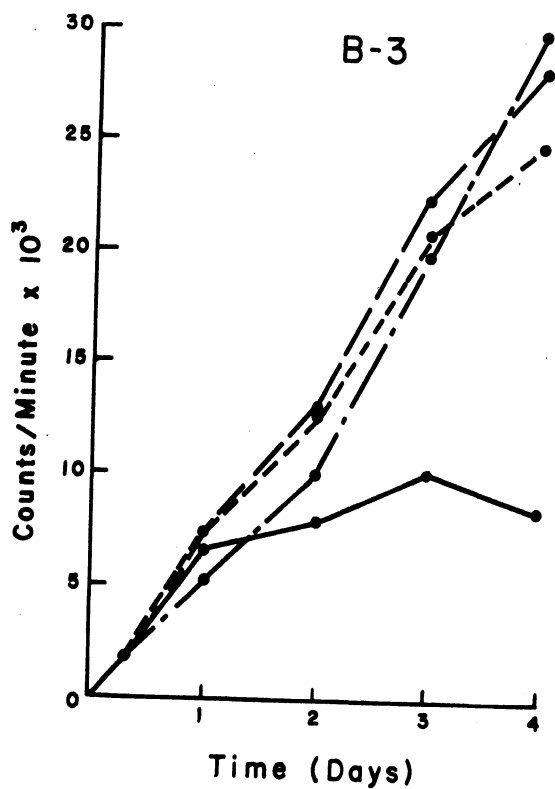
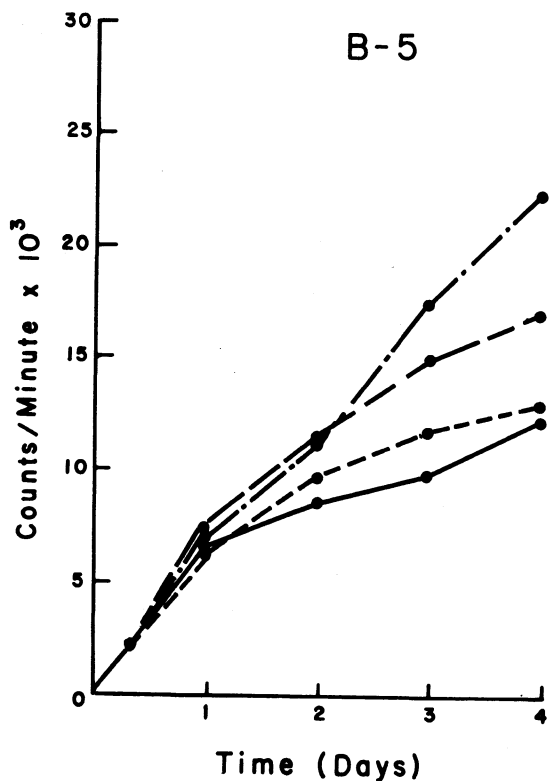
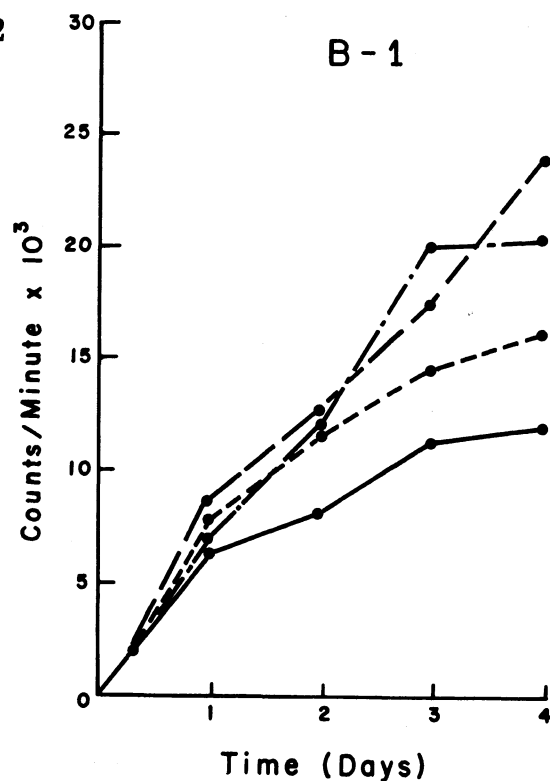


Fig. 25. Uptake of carbon-14 (counts per minute) by Lake Michigan phytoplankton when subjected to various concentrations of Fuller's Earth sediment extract.



- Control
- - -●- - - 5 ml Sediment Extract
- · - · -●- · - · 10 ml Sediment Extract
- - · - ·●- · - · 25 ml Sediment Extract

Fig. 26. Uptake of carbon-14 (counts per minute) by Lake Michigan phytoplankton when subjected to various concentrations of Buffalo, N. Y., harbor sediment extract (stations B-1, B-3, B-5).

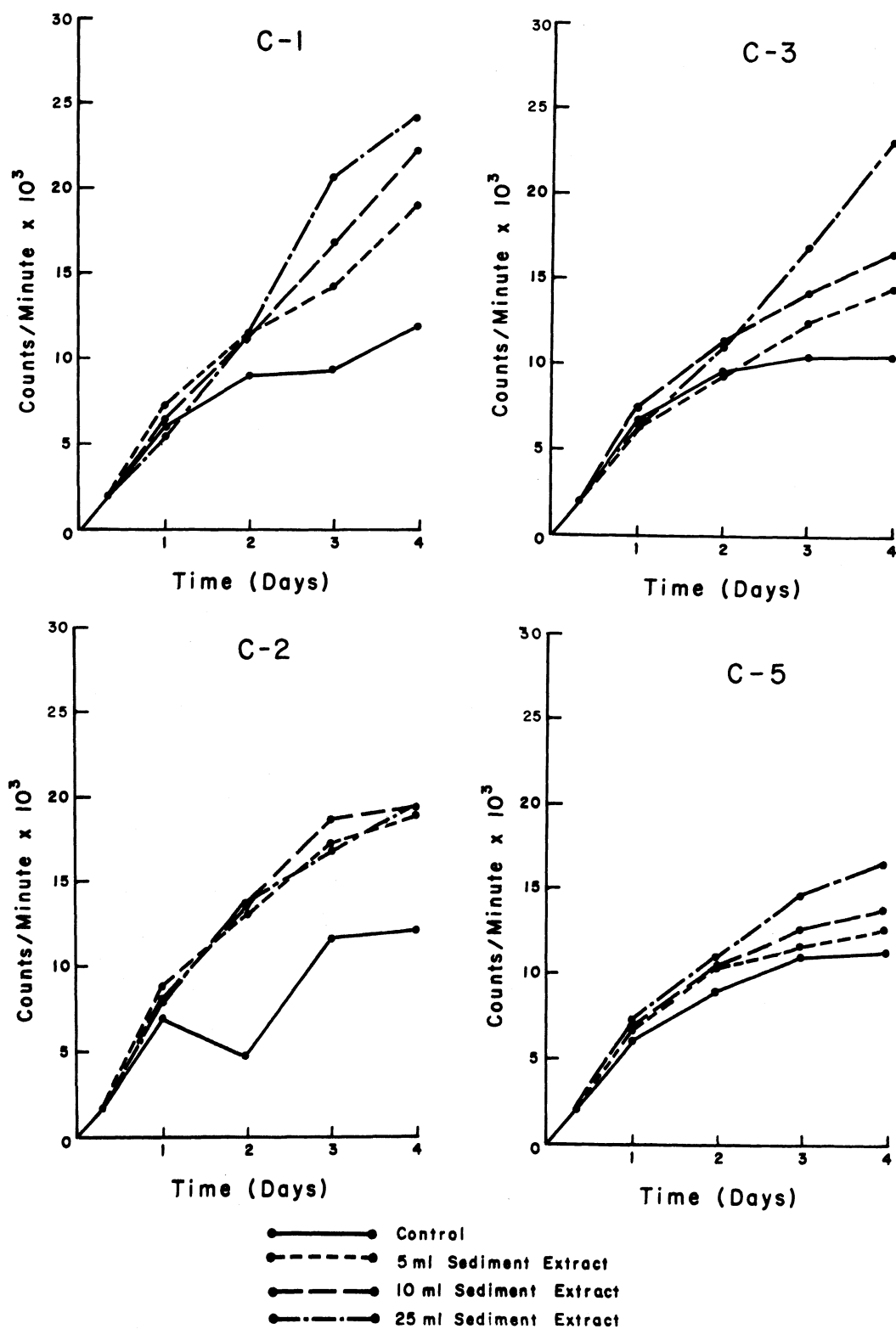


Fig. 27. Uptake of carbon-14 (counts per minute) by Lake Michigan phytoplankton when subjected to various concentrations of Calumet, Ill., harbor sediment extracts (stations C-1, C-2, C-3, C-5).

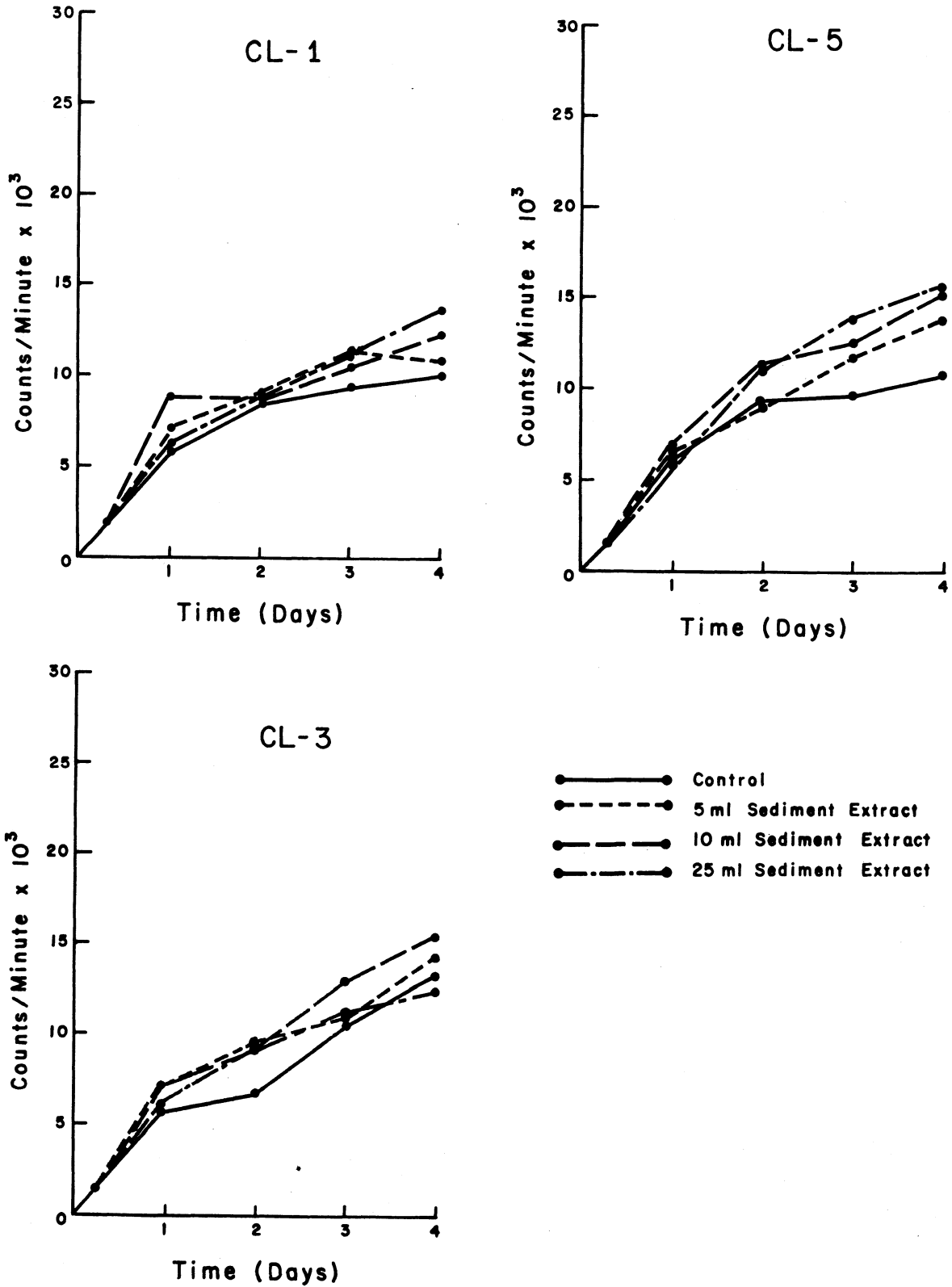


Fig. 28. Uptake of carbon-14 (counts per minute) by Lake Michigan phytoplankton when subjected to various concentrations of Cleveland, Ohio, harbor sediment extracts (stations CL-1, CL-3, CL-5).

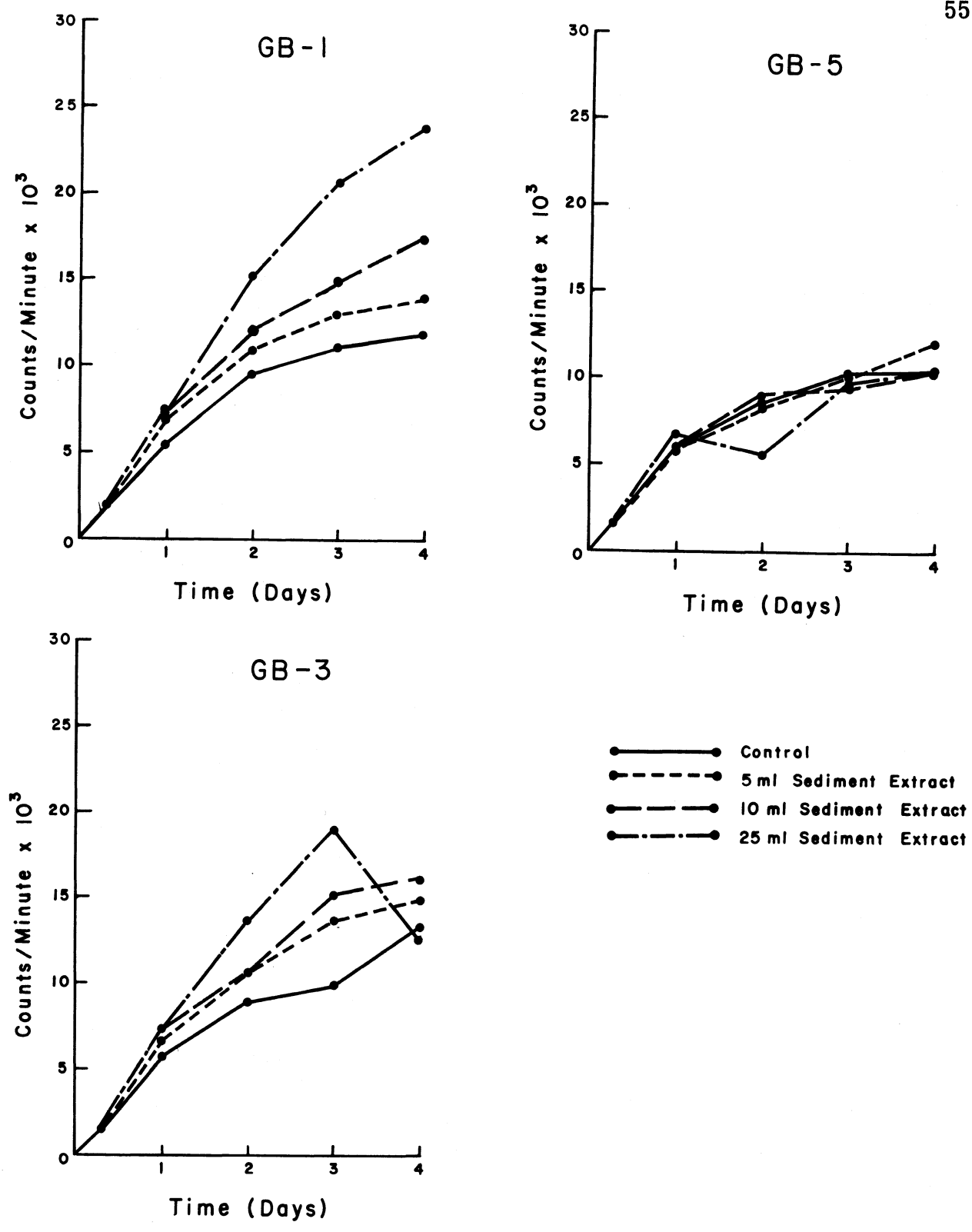


Fig. 29. Uptake of carbon-14 (counts per minute) by Lake Michigan phytoplankton when subjected to various concentrations of Green Bay, Wis., harbor sediment extracts (stations GB-1, GB-3, GB-5).

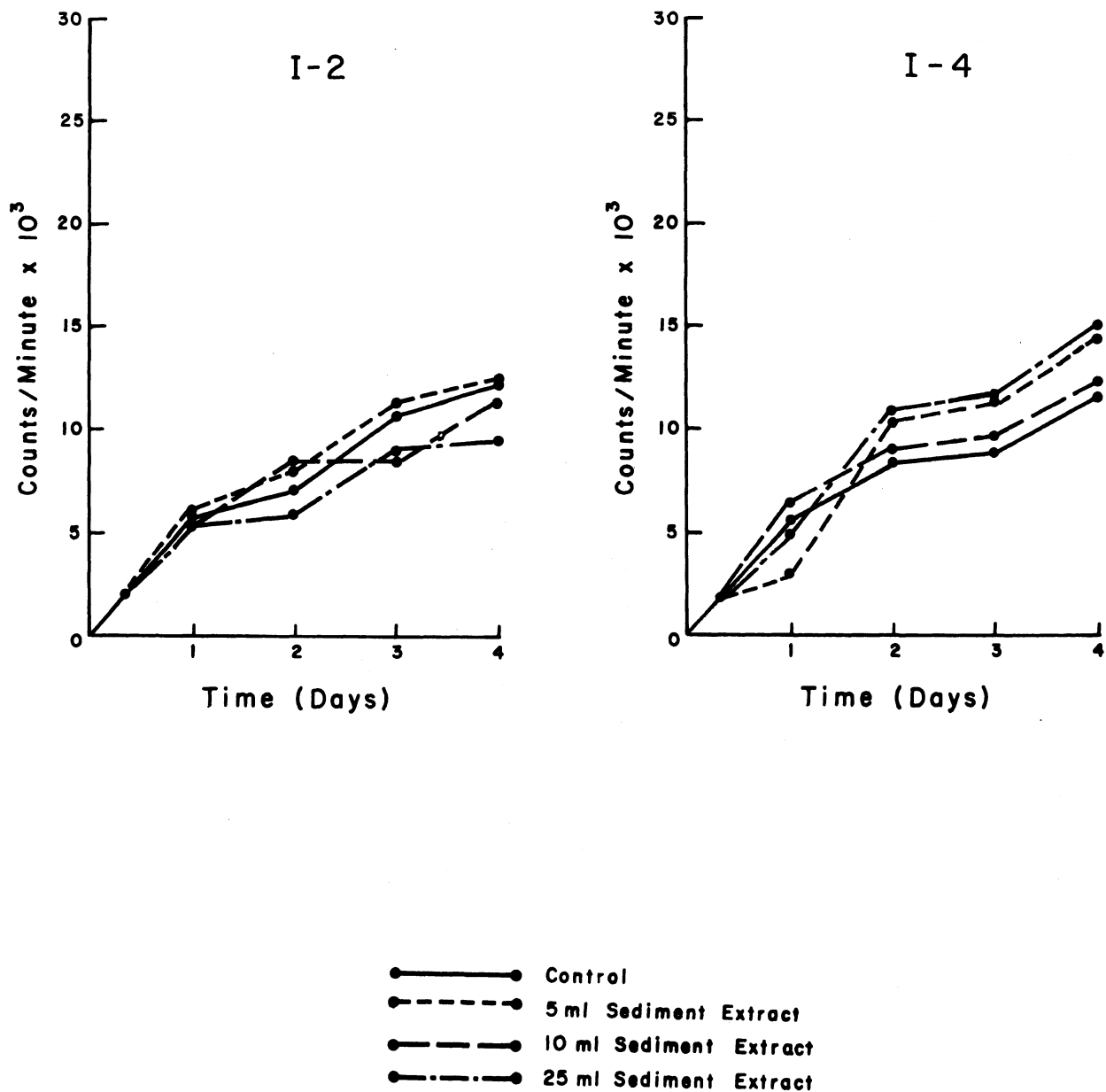


Fig. 30. Uptake of carbon-14 (counts per minute) by Lake Michigan phytoplankton when subjected to various concentrations of Indiana harbor sediment extracts (stations I-2, I-4).

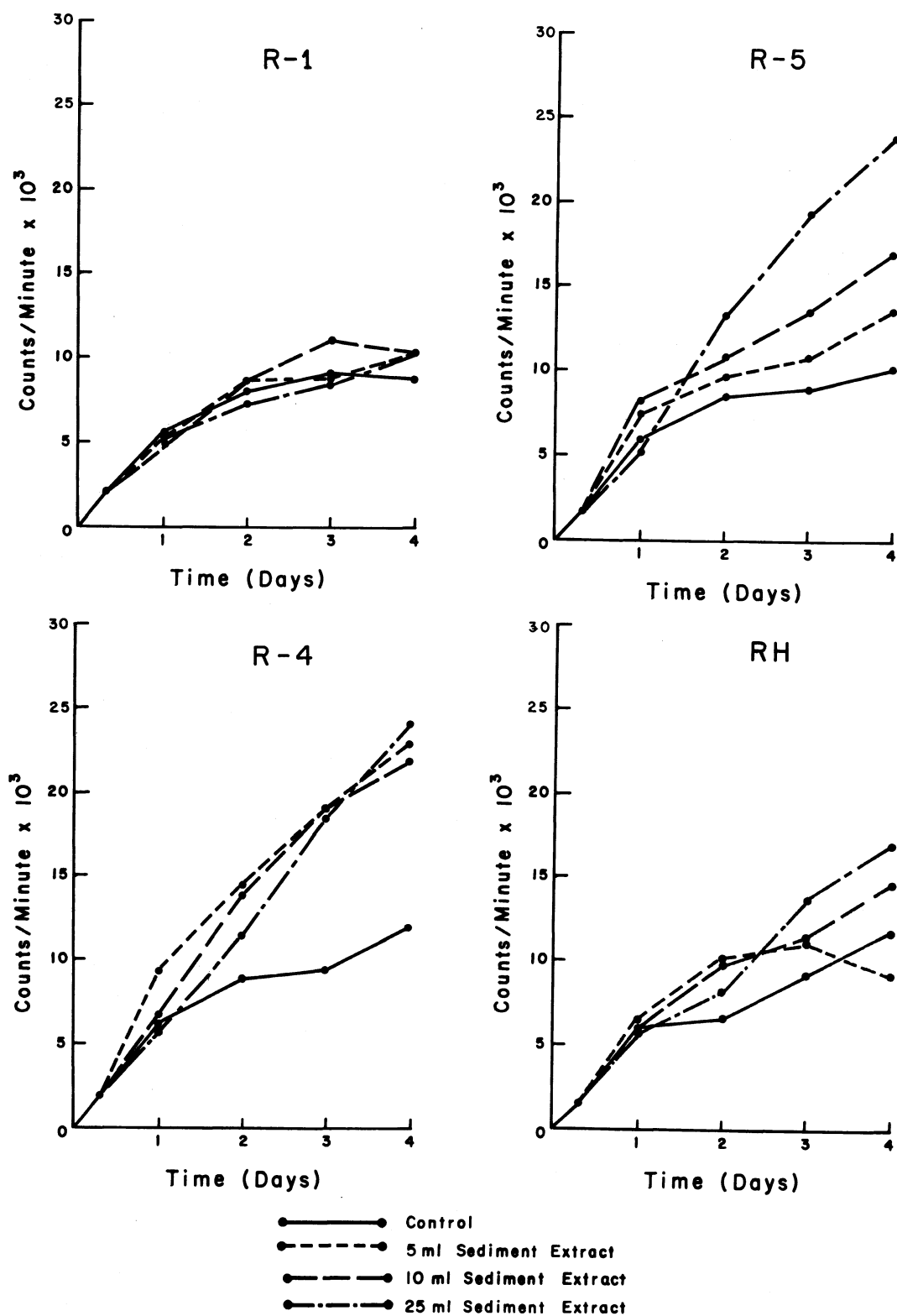


Fig. 31. Uptake of carbon-14 (counts per minute) by Lake Michigan phytoplankton when subjected to various concentrations of Rouge River (Detroit, Mich.) harbor sediment extracts (stations R-1, R-4, R-5, RH (hopper dredge)).

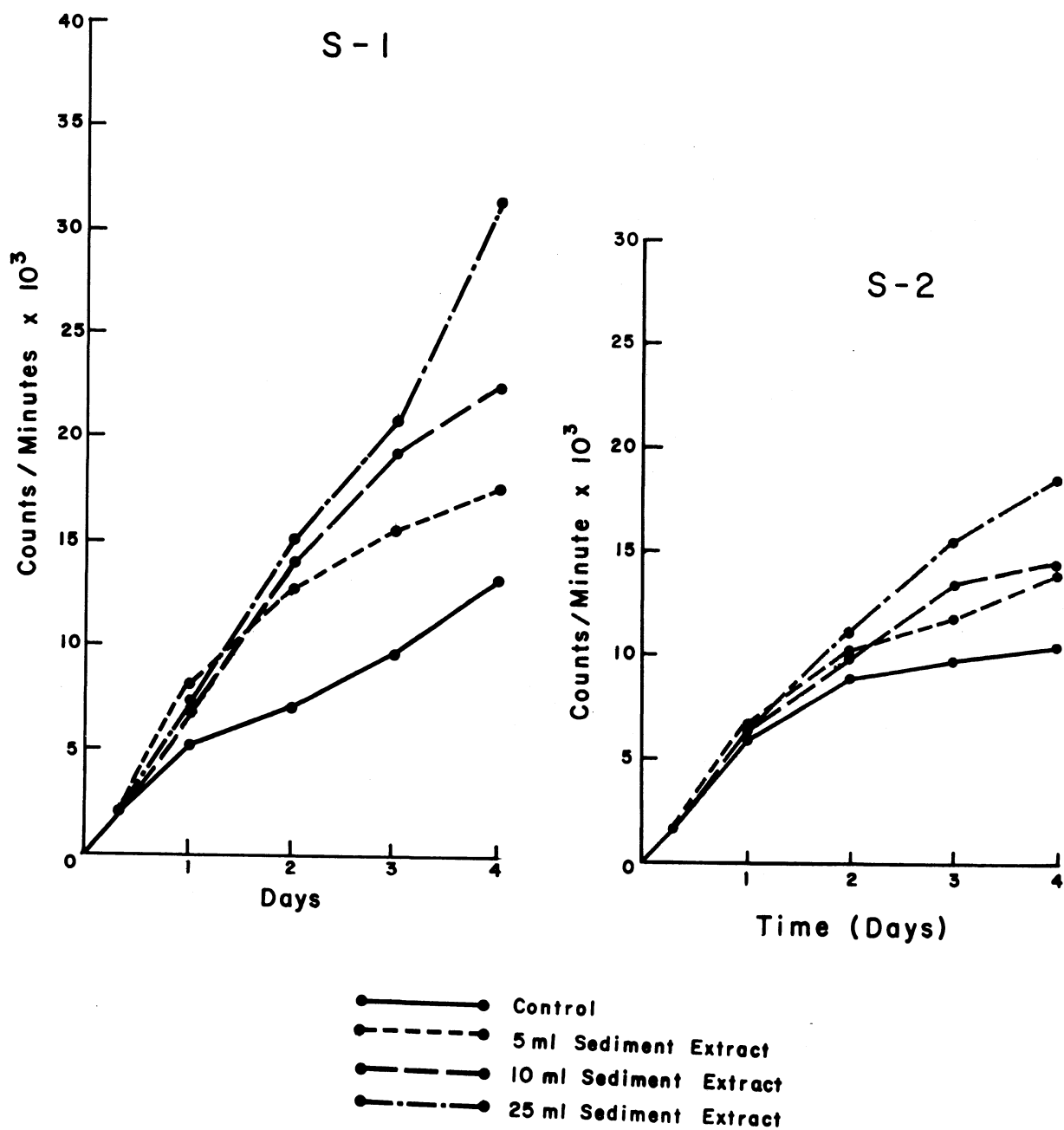


Fig. 32. Uptake of carbon-14 (counts per minute) by Lake Michigan phytoplankton when subjected to various concentrations of Great Sodus Bay, N. Y., harbor sediment extracts (stations S-1, S-2).

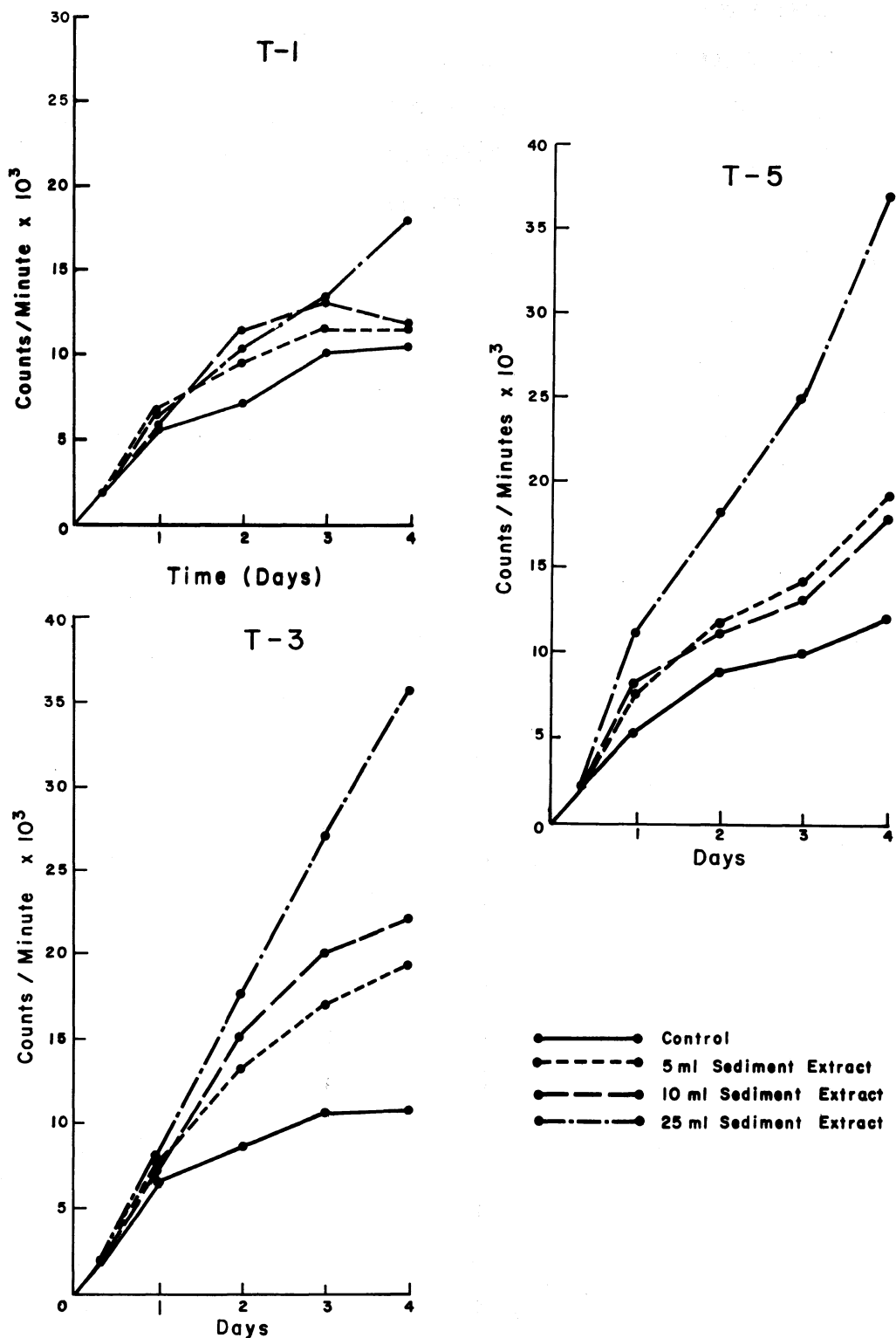


Fig. 33. Uptake of carbon-14 (counts per minute) by Lake Michigan phytoplankton when subjected to various concentrations of Toledo, Ohio, harbor sediment extracts (stations T-1, T-3, T-5).

uptake were those with sediments high in phosphorus, but the tests with little or no uptake were also those with sediments having ample nutrients. In this latter case, some sediments may have toxic substances which would suppress growth. It should be emphasized that algal nutrition is very complex. It is well established that too much of some nutrient as well as too little can be limiting (Fogg, 1966). Furthermore, results of laboratory studies of algal nutrition can be applied to the natural situation only with extreme caution.

#### Fuller's Earth

No significant uptake of carbon-14 occurred in the Fuller's Earth extract samples relative to the control (Fig. 25).

#### Buffalo

The uptake of carbon-14 in the tests with the 3 sediments was above the average ( $20 \text{ counts} \times 10^3/\text{minute}$ ) for all sediments (Fig. 26). Phosphate concentrations were above the average ( $0.43 \text{ mg/gm}$ ) for all the sediments. Ammonia and organic nitrogen concentrations were below the average concentrations of  $0.109$  and  $2.20 \text{ mg/gm}$  (Table 3).

#### Calumet

The uptake of carbon-14 was above average for all four of the Calumet samples (Fig. 27). Phosphate concentrations were above average and ammonia and organic nitrogen below average.

#### Cleveland

The uptake of carbon-14 was increased only slightly by the addition of Cleveland sediments (Fig. 28). Phosphate, ammonia, and organic nitrogen concentrations were above average.

#### Green Bay

The GB-1 sample resulted in the greatest uptake of carbon-14 of the three Green Bay sediments (Fig. 29). It had the highest organic

nitrogen content and an ammonia content twice that of the GB-5 sample. Carbon-14 uptake in the GB-5 test was insignificant. The phosphorus content of the GB-5 sediment was 2 to 3 times lower than that of GB-3 and GB-1 (Table 3).

#### Indiana Harbor

No significant uptake of carbon-14 occurred in tests with I-2 and I-4 sediments (Fig. 30).

#### Rouge River

No significant increase occurred in the uptake of carbon-14 in the test with R-1 sediment; the increase was significant for the R-4 and R-5 samples and about average for all the sediments (Fig. 31). Concentrations of ammonia, organic nitrogen, and phosphorus were near the average for all the harbors. Little or no uptake was noted in the Rouge River hopper dredge sample (Fig. 31).

#### Great Sodus Bay

The bioassays with sediment S-1 had a high uptake of carbon-14 whereas only a slight uptake occurred with the S-2 sample (Fig. 32). The ammonia content of the sediments was much lower than the average. Organic nitrogen and phosphorus concentrations were about average (Table 3).

#### Toledo

The bioassays with the T-3 and T-5 sediments resulted in the highest uptake of carbon-14 of any of the samples (Fig. 33). Uptake in the T-1 test was about average. The T-3 and T-5 samples had the highest ammonia content of any of the sediments and higher than average concentrations of organic nitrogen and phosphorus (Table 3).

### E. Bioassay of Sediments Using *Daphnia pulex* and Zooplankton

These tests were designed to determine whether the sediments were toxic to planktonic animals. The results are useful in giving some insight to the possible direct effects of open-lake disposal on the zooplankton.

#### 1. *Daphnia pulex*

This small planktonic crustacean is easily cultured in the laboratory, and this species and related species frequently have been used in bioassays of toxic substances (Anderson, 1944).

The mortalities in the control tests were high ( $42\% \pm 16$  standard deviation, range 26 to 58%). The reason for this high mortality is unknown. A mortality of 60% (average mortality in the control plus the standard deviation) or more is considered as significant in the following presentation of results.

An attempt was made to relate the mortality of *Daphnia* to some chemical characteristics of the sediments. When the results from individual harbors were examined, it appeared that a significant mortality of *Daphnia* occurred when a sediment with a high COD (Table 3) was introduced into their environment. This relationship is not clear cut when the results of the tests from all the harbors are lumped together. The COD of the sediments is not considered as the toxic factor. The conclusion is that a high COD may indicate a sediment that will be toxic.

#### Buffalo

Significant mortality occurred in all three dilutions of the sediments, except for the sample from B-1 (Fig. 34). Sample B-1 had a COD which was 2 to 3 times less than the others (Table 3).

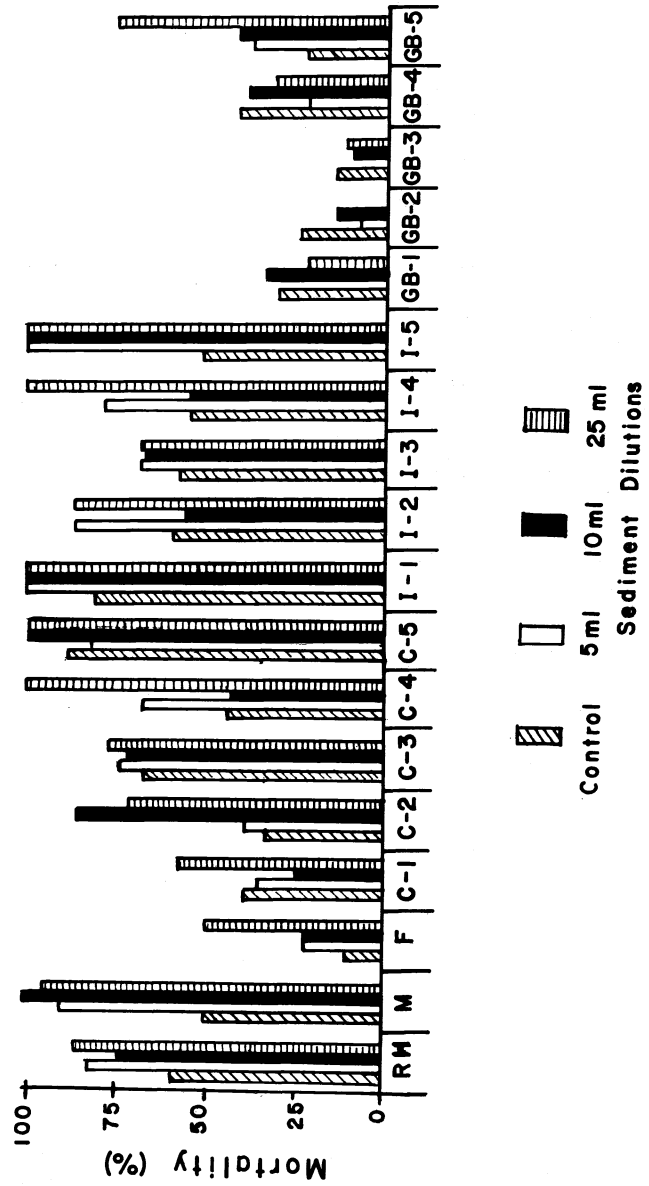
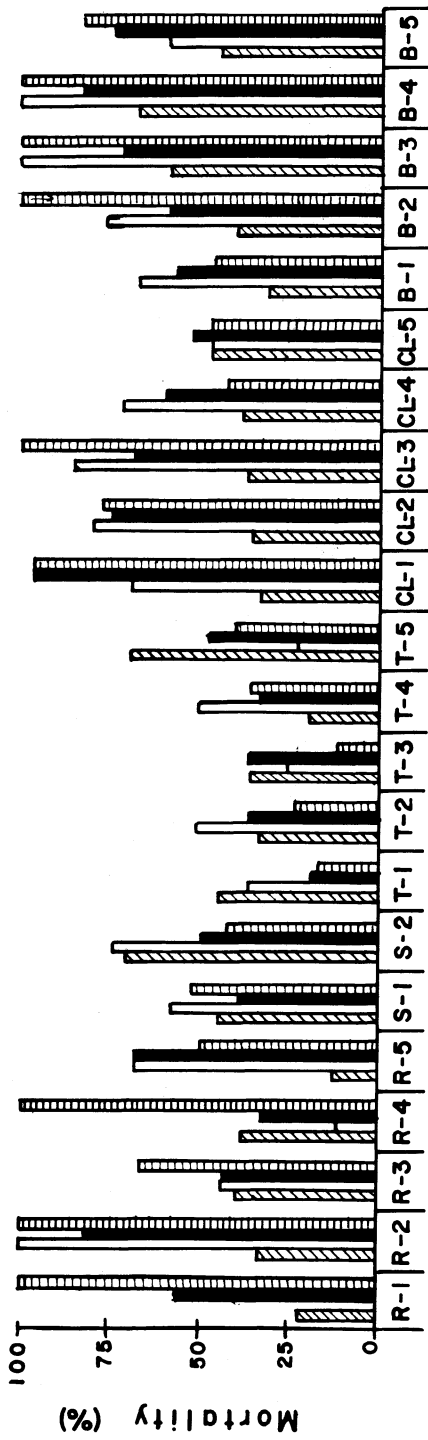


Fig. 34. Mortality of *Daphnia pulex* in various concentrations of harbor sediments (B, C, CL, GB, I, M, R, RH, S, and T) and Fuller's Earth (F).

### Calumet

The mortalities in the tests with the river sediments were significant, except for the 5 ml dilution from C-2 and the 10 ml dilution from C-4. No significant mortality occurred in the outer harbor (C-1) sample. The river sediments all had high COD's (Table 3).

### Cleveland

Mortalities in the tests with river sediments were significant. Mortality in the sample from the CL-5 outer harbor station was not significant, whereas the results from the other outer harbor sample was approaching a level of significant mortality (45.4 to 73.3%). The river samples had higher COD's than the harbor samples (Table 3).

### Great Sodus Bay

Mortality was not significant in either of the harbor samples. Both had low COD's.

### Green Bay

Mortalities were not significant in any of the tests, except for the 25 ml dilution of the sample from GB-5.

### Indiana Harbor

Mortalities were significant in all the harbor sediments. The highest mortalities (100%) occurred in the tests with the I-4 and I-5 sediments. These sediments had higher COD's than the other samples (Table 3).

### Rouge River

Significant mortalities occurred in the tests with the greatest amount of sediment, except in the R-5 sample. The percentage mortality was highest in the R-2 sample. Mortalities in the R-3, R-5 and in the 5 ml and 10 ml dilutions of R-4 samples were either insignificant or approaching a significant level.

## Toledo

No significant mortality occurred in any of the tests. These samples had low COD's.

## 2. Zooplankton

Results of the tests with naturally occurring zooplankton from one of the Great Lakes (Michigan) represent a more valid test of the possible effect of open-lake disposal on planktonic animals than the tests with Daphnia in culture. The results were closely similar to those of the previous test. Mortalities in the control tests were  $33\% \pm 13$  (standard deviation). Mortality of 50% or more was considered as significant in this experiment.

## Buffalo

Significant mortalities occurred in all the sediments, with 100% mortalities in the river sediments (Fig. 35).

## Calumet

Mortality in C-1 & C-5 samples was insignificant. Significant mortalities occurred in all dilutions of the C-3 sample and in the 25 ml dilution of C-2 and the 10 and 25 ml dilutions of C-4.

## Cleveland

Mortalities in the tests of river sediments were greater than 50%, but mortalities in the tests with outer harbor sediments were at or slightly below the 50% level of significance.

## Great Sodus Bay

No significant mortality occurred.

## Green Bay

Mortality was not significant in any tests, except those from GB-1.

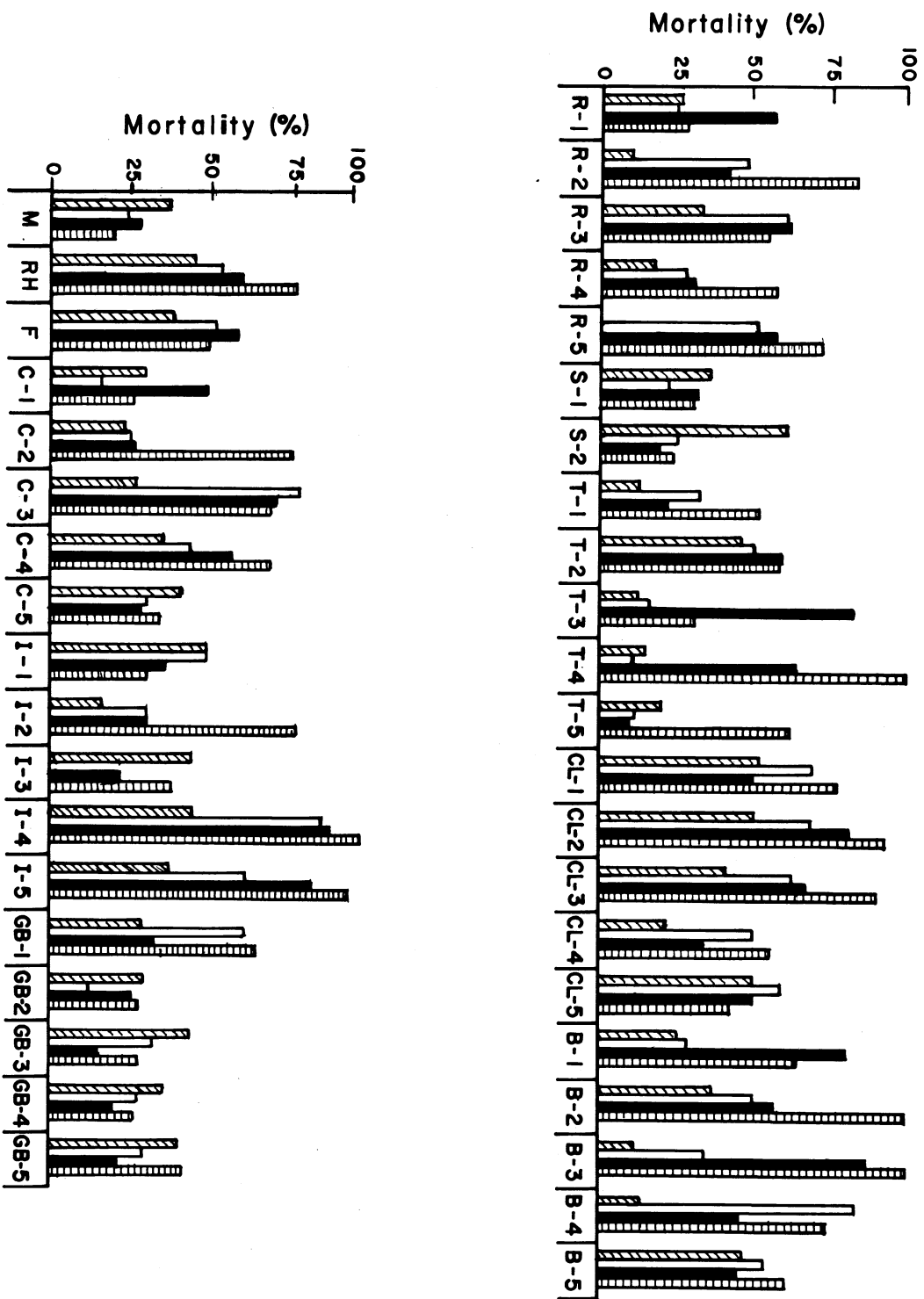


Fig. 35. Mortality of zooplankton in various concentrations of harbor sediments (B, C, CL, GB, I, M, R, RH, S, and T) and Fuller's Earth (F).

### Indiana Harbor

Mortalities in the test of samples from I-1, I-2, and I-3 were not significant, except for the 25 ml dilution of I-3. Mortalities were high in the I-4 and I-5 samples. The COD's of these sediments were much higher than in the other samples (Table 3).

### Rouge River

Significant mortalities occurred in tests of each sediment with greatest mortalities occurring in the least diluted sediment samples. Mortalities were less than 50% in the most diluted sediment, except for the 5 ml dilution of R-3.

### Toledo

Significant mortalities occurred in all the sediments when they were in the greatest concentration (25 ml), except for sample T-3 with the greatest mortality in the 10 ml test. All three dilutions of the T-2 sediment resulted in significant deaths of Daphnia.

### F. Bioassay of Sediments Using the Benthic Alga, Cladophora

Several experiments were attempted to determine the effects of various sediments on Cladophora. Most of these failed because of lack of growth and eventual loss of pigment and death under the laboratory conditions. The experiments, where the sediments were introduced into the experimental flasks containing Cladophora, failed because sediment particles clung to the algal filaments and the algae deteriorated within 48 hrs. These experiments showed that Cladophora would not grow in direct contact with the sediments, at least under laboratory conditions, and the experiments failed because no quantitative measurements could be made. We had originally intended to determine growth by changes in the weight of the pieces of Cladophora.

Active growth of Cladophora was finally realized by adding soil extract to the culture medium and by making extracts of the sediments for the bioassays. We intended to determine the growth of Cladophora by measuring the increase in length, and pieces of algae were selected that did not have any branching. During the experiment, many pieces of Cladophora did increase in length, but branching also occurred. Consequently, it was not possible to deal with the results quantitatively and the results can be expressed only subjectively in terms of whether considerable growth did or did not occur. Even an interpretation on this basis is doubtful in view of the results from the control tests where the mean increase in length was 2.1 mm with a range of 0 to 6 mm. Furthermore, Fuller's Earth was also included as a control sediment, and algal growth was almost as great as with many of the harbor sediments.

Some of the sediments apparently stimulated the growth of Cladophora, if branching and an increase in length of greater than 5 mm are accepted as an indication of significant growth. One sample from Buffalo Harbor (B-4), three sediments from Cleveland (CL-1, CL-4, and CL-5), three sediments from Indiana Harbor (I-3, I-4, I-5) and two from Toledo (T-2, T-3) stimulated the growth of Cladophora. Growth was greater in the 10 ml dilution of sediments for eight of the samples than in 25 ml dilution. In the other seven samples, growth was greatest in the 25 ml dilution.

It appears that the sediment extracts did increase the growth of Cladophora, but the results are inconclusive. Cladophora was not easy to culture in the laboratory, and it is difficult to make a precise measurement of its growth. Consequently, this plant was not especially suitable for our bioassay methods.

TABLE 4. Response of *Cladophora* to various sediments. (Samples are indicated by a letter designating the harbor, a small number for the sampling location, and a larger number (10 or 25) for the amount of sediment extract (ml) added to the culture.)

Sample	Initial length (mm)	Final length (mm)	Increase in length (mm)	Other Observations
B-1-10	11	14	3	Moderate lateral branching
B-1-25	10	15	5	Moderate lateral branching
B-2-10	10.5	15	4.5	Moderate lateral branching, few blank cells present
B-2-25	10	12	2	Moderate lateral branching
B-3-10	10	12	2	Heavy lateral branching
B-3-25	11	13	2	Blank cells present, epiphytic diatoms
B-4-10	10	16	6	Moderate lateral branching, epiphytic diatoms
B-4-25	10	15	5	Moderate lateral branching, bright green in color
B-5-10	10	15	5	Moderate lateral branching, epiphytic diatoms
B-5-25	13	15	2	Moderate lateral branching, bright green in color
C-1-10	10	10	0	Moderate lateral branching, epiphytic diatoms
C-1-25	5	7	2	Moderate lateral branching, blank cells present
C-2-10	11	15	4	Moderate lateral branching, epiphytic diatoms
C-2-25	11.5	15	4.5	Moderate lateral branching, bright green in color, epiphytic diatoms
C-3-10	11	11	0	Moderate lateral branching, few blank cells
C-3-25	11	13	2	Moderate lateral branching, bright green in color
C-4-10	11	11	0	Heavy lateral branching, epiphytic diatoms
C-4-25	10	15	5	Moderate lateral branching

TABLE 4, continued.

Sample	Initial length (mm)	Final length (mm)	Increase in length (mm)	Other Observations
C-5-10	9	11	2	Heavy lateral branching
C-5-25	10	13	3	Moderate lateral branching, epiphytic diatoms
CL-1-10	10	16	6	Sparse lateral branching, a few blank cells
CL-1-25	9	10	1	Heavy lateral branching
CL-2-10	11	15	4	Sparse lateral branching, epiphytic diatoms
CL-2-25	7	7.5	0.5	Blank cells common, sparse lateral branching
CL-3-10	8	9	1	No lateral branching, blank cells common
CL-3-25	10	11	1	Moderate lateral branching, blank cells
CL-4-10	11	23	12	Sparse lateral branching
CL-4-25	12	20	8	Moderate lateral branching, epiphytic diatoms
CL-5-25	8	18	10	Moderate lateral branching, a few blank cells
GB-1-10	10	11	1	Heavy lateral branching
GB-1-25	10	10	0	Moderate lateral branching
GB-2-10	9	12	3	Moderate lateral branching, many epiphytic diatoms
GB-2-25	9	13	4	Sparse lateral branching
GB-3-10	11	14	3	Sparse lateral branching, epiphytic diatoms
GB-3-25	15	16	1	Sparse lateral branching, bright green in color
GB-4-10	8	12	4	Covered with epiphytes, blank cells present
GB-4-25	11	13	2	Covered with epiphytes
GB-5-10	9	12	3	Very heavy lateral branching
GB-5-25	11	16	5	Heavy lateral branching, epiphytic diatoms

I-1-10	10	13	3	Sparse lateral branching, light green in color
I-1-25	11	12	1	Moderate lateral branching, epiphytic diatoms
I-2-10	10	13	3	Bright green, heavy epiphytic diatoms
I-2-25	8	8	0	Moderate lateral branching
I-3-10	12	19	7	Heavy lateral branching
I-3-25	8	9	1	Heavy lateral branching, some blank cells
I-4-10	11	14	3	Moderate lateral branching
I-4-25	12	20	8	Heavy lateral branching
I-5-10	11.5	25	13.5	Moderate lateral branching
I-5-25	12	15	3	Moderate lateral branching, a few blank cells
M-10	6	7	1	Moderate lateral branching
M-25	11	13	2	Sparse lateral branching, a few blank cells
R-1-10	10.5	13	2.5	Moderate lateral branching, epiphytic diatoms, a few blank cells
R-1-25	9	10	1	Heavy lateral branching, a few blank cells
R-2-10	7.5	8	0.5	Moderate lateral branching, diatoms and other epiphytes
R-2-25	11	11	0	A few blank cells, sparse lateral branching
R-3-10	7	8	1	Moderate lateral branching, epiphytic diatoms
R-3-25	6	7	1	Blank cells, epiphytic diatoms
R-4-10	11	14	3	Moderate lateral branching, epiphytic diatoms
R-4-25	8	9	1	Heavy lateral branching, epiphytic diatoms
R-5-10	9.5	12	2.5	Heavy lateral branching, epiphytic diatoms
R-5-25	9	12	3	Moderate lateral branching, bright green in color

TABLE 4, continued.

Sample	Initial length (mm)	Final length (mm)	Increase in length (mm)	Other Observations
S-1-10	11	14	3	Lateral branching, epiphytic diatoms
S-1-25	9	10	1	Lateral branching
S-2-10	7	7	0	Sparse lateral branching, epiphytic diatoms
S-2-25	11	12	1	Heavy lateral branching, epiphytic diatoms
T-1-10	10	13	3	Heavy lateral branching, epiphytic <u>Vorticella</u> sp.
T-1-25	12	13	1	No lateral branching
T-2-10	10	10.5	0.5	Moderate lateral branching, many blank cells
T-2-25	10	20	10	Heavy lateral branching
T-3-10	12	18	6	Little lateral branching, blank cells present
T-3-25	14	15	1	No lateral branching, many blank cells, thallus in poor condition
T-4-10	10	12	2	Many blank cells, no lateral branching
T-4-25	10	10	0	Blank cells common, no lateral branching
T-5-10	9	9	0	Moderate lateral branching, a few blank cells, epiphytes
T-5-25	10.5	13	2.5	Moderate lateral branching
F-10	11.5	15	3.5	Moderate lateral branching, bright green in color
F-25	10	20	10	Profuse lateral branching

Control 1	7	10	3	Blank cells present
Control 2	10	13	3	Light green in color
Control 3	8	13	5	Heavy lateral branching
Control 4	10	16	6	Moderate lateral branching
Control 5	10	12	2	Heavy lateral branching
Control 6	6	none present		
Control 7	9	9	0	Moderate lateral branching, blank cells present
Control 8	7.5	9.5	2	Heavy lateral branching
Control 9	11	11	0	Many blank cells, epiphytic diatoms
Control 10	9	9	0	Many blank cells, thallus in poor condition

## V. CONCLUSIONS

The harbor sediments fit into five categories in terms of their effect on the test organisms.

1. Some sediments were toxic, were avoided by benthic animals, and did not stimulate growth of phytoplankton or Cladophora. These sediments were Buffalo (B-2), Calumet (C-4), Cleveland (CL-2, CL-3), Indiana Harbor (I-1, I-2), Rouge River (R-1, R-2, R-3), and Toledo (T-4).

2. A large number of sediments were toxic to the test animals, but they stimulated the growth of phytoplankton or Cladophora. Sediments in this category came from Buffalo (B-1, B-3, B-4 and B-5), Calumet (C-3, C-5), Cleveland (CL-1), Indiana Harbor (I-3, I-4, I-5), and Rouge River (R-4, R-5).

3. Some sediments were only somewhat toxic, i. e. , killed some test animals but did not affect others; they were avoided by Pontoporeia but not by midge larvae; and they stimulated growth of phytoplankton or Cladophora. These sediments were Calumet (C-2), Cleveland (CL-4, CL-5), Great Sodus Bay (S-1), Green Bay (GB-1), and Toledo (T-1, T-2, T-3, T-5).

4. Three sediments from Green Bay (GB-2, GB-4, GB-5) were not especially toxic and did not stimulate growth of the algae.

5. Three sediments were not toxic and they stimulated growth of phytoplankton but not the growth of Cladophora. These sediments were Calumet (C-1), Great Sodus Bay (S-2), and Green Bay (GB-3).

The five categories in the summary form a natural sequence ranging from the least suitable sediments for life, i. e. , those that would certainly be considered as pollutants, to those that form a suitable environment for aquatic organisms. All of the Buffalo sediments, the three inner harbor samples from Calumet, the three river samples from

Cleveland, all of the Indiana Harbor sediments, all of the Rouge River sediments, and the sample collected near the outfall of the Toledo sewage treatment plant (T-4) all fall into the first two categories. The least objectionable sediments appear to be from the outer harbor of Calumet (C-1), Great Sodus Bay (S-2), and Green Bay (GB-3). The sediment from the outer harbor of Calumet was one of the least toxic sediments. This harbor is different from the others, however, since Lake Michigan water flows into the Calumet River. Consequently, there is a continuous influx of high quality water into the outer harbor. This situation also explains the somewhat better condition of the sediment from the mouth of the Calumet River (C-2) in comparison with the inner harbor sediments.

The sediments from Green Bay and the Fox River, which is considered to be badly polluted, were in general of much better quality than those from the other harbors. It is very likely that the samples, which were collected for this study, do not represent the usual conditions, since they were collected after dredging was completed. Furthermore, this was new work to deepen the channel and not maintenance dredging. Nevertheless, if this is the situation, the results indicate improvement in the harbor environment due to dredging.

Although harbors are very complex environments from an ecologist's viewpoint, it would be desirable to relate the results from these biological tests to various physical and chemical parameters of the harbor sediment. We found no direct correlation between any single physical or chemical parameter and trends in the biological test results. However, when several chemical parameters were examined collectively, a possible trend could be discerned. We observed that chemical oxygen demand (COD), volatile solids, phosphate-phosphorus and ammonia increased progressively from harbor category 1 through category 5 as established by the biological tests (Fig. 36). Further research may reveal definite relationships between some of the chemical parameters and mortality of organisms. Consequently, biological testing should be

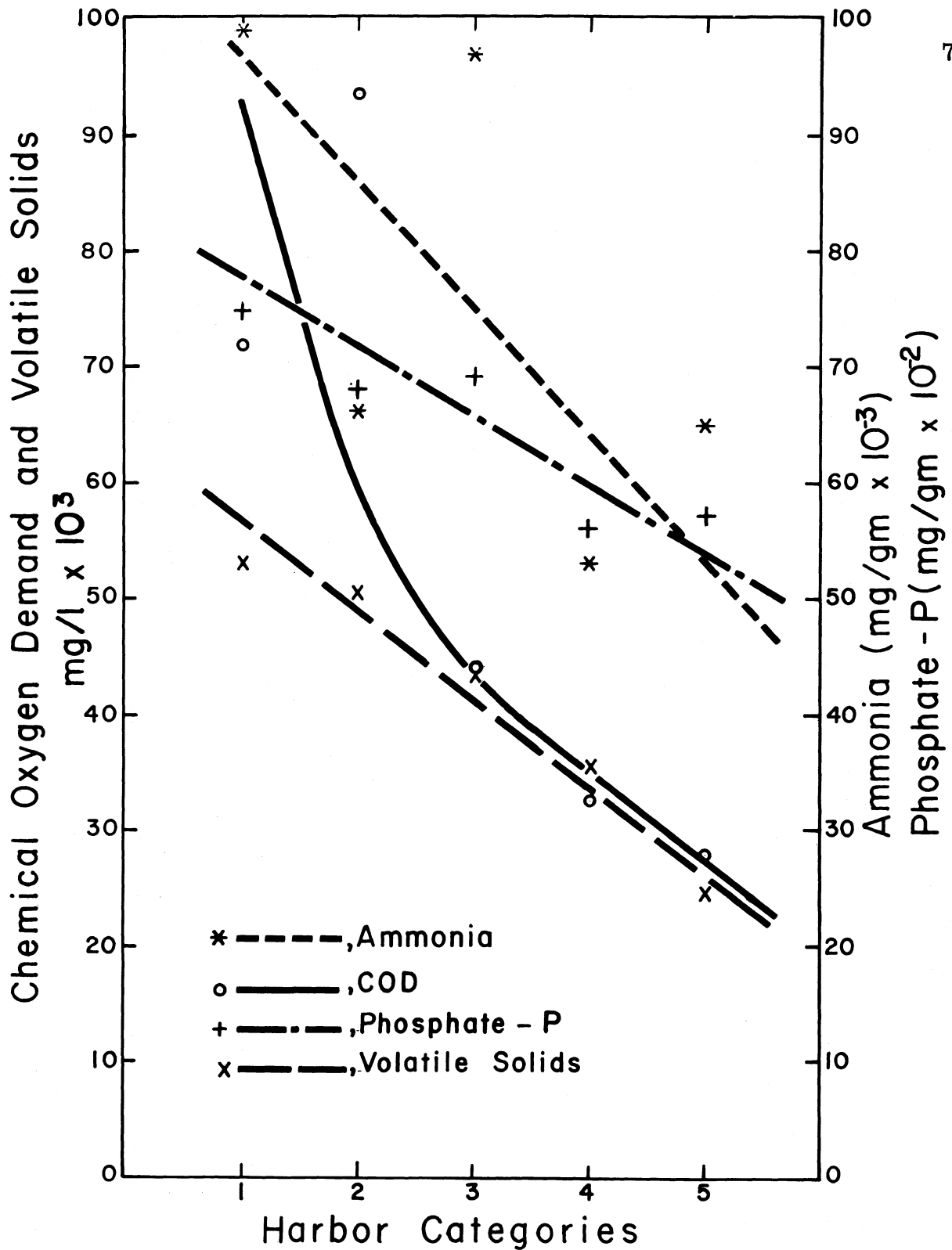


Fig. 36. Possible relationship between results of the biological tests and certain chemical characteristics of the sediments. The harbor sediments fit into 5 categories in terms of their effects on organisms (see text, p. 75), ranging from least suitable for life (Category 1) to most suitable for life (Category 5).

supplemented with chemical data such as BOD, COD, algal nutrients, volatile solids, oil and grease, and others, to assist in the interpretation of the biological results.

The study has accomplished the original objectives. Simple bioassay and selectivity experiments were useful in indicating some direct effects of harbor sediments on benthos and plankton. The results are only preliminary, however. It would have been better to sample several additional stations in each harbor, and to have composite samples from each station. Also, all biological tests must be run in duplicate if not triplicate before definite conclusions can be drawn from the results. Time allowed for the contract work did not permit further testing.

The greatest amount of useful information was obtained from viability experiments, sediment selectivity tests, and the phytoplankton bioassays using carbon-14. These methods offer the greatest potential for further studies of harbor sediments and biota.

Future studies on viability of benthic organisms in harbor sediments should investigate whether mortality of test organisms is actually due to toxic substances in the sediments or to a high oxygen demand of the sediments. Large capacity test containers should be used, and oxygen content monitored by micro-winkler titrations or with a self-stirring oxygen electrode. In addition, standard biological oxygen demand (BOD) tests may be desirable (APHA, 1965).

When using small, delicate invertebrates, such as zooplankton, for bioassays, some mortality occurs from capture, transport, and transfer operations. Therefore, it is very important to run many controls. Traditional fish bioassay measurements, such as TLM 50 (APHA, 1965) often cannot be directly applied to small invertebrates. In our tests, we took the average mortality ( $\bar{m}$ ) in all controls in each experiment and added the standard deviation (S.D.) of the mortality. Mortality was considered significant when above ( $\bar{m} + \text{S.D.}$ ).

The tests in which sediment was introduced into algal cultures and measurements of change were based on optical density did give some insight into the possible direct effect of harbor sediment on

planktonic algae. If tests of this nature are conducted again, the degree of clumping of sediment particles and algal cells must be determined as well as whether any change in growth actually occurs. Direct observation must be employed as a check against optical density measurements. Since sediment particles will interfere with microscope counting methods, other methods of enumeration should be explored. Density gradient separation (Lammers, 1962) or fluorescent techniques commonly used in bacteriology (Gurr, 1956) offer possibilities for algal culture and phytoplankton work.

Bioassays using Cladophora were especially difficult to interpret. This alga is difficult to grow in culture unless ample nutrients are provided including soil extract. In addition, the morphology of the thallus changes in culture from the wild parental type (Bellis, 1968). Future bioassays using Cladophora possibly should employ gentle constant agitation of the culture flasks or a constant flow of water in an artificial stream.

Perhaps the most important aspect concerning this study is that basic data on the direct effects of various pollution factors on biota such as presented here are sadly lacking. Routine pollution surveys and monitoring must be supplemented with experimental procedures. The methods used in this study are simple. They can easily be modified to answer other questions of concern in water pollution biology.

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