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thesis entitled

ENVIRONMENTAL FACTORS AFFECTING  
THE DISTRIBUTION AND ABUNDANCE OF CHIRONOMIDAE  
(DIPTERA) IN TWO SEWAGE OXIDATION LAKES  
presented by

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has been accepted towards fulfillment  
of the requirements for

M.S. degree in Entomology

A handwritten signature in cursive script, reading "Richard W. Merritt". The signature is written in dark ink and is positioned above the title "Major professor".

Major professor

Date 3 May 1978

ENVIRONMENTAL FACTORS AFFECTING  
THE DISTRIBUTION AND ABUNDANCE OF CHIRONOMIDAE  
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By

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## ABSTRACT

### ENVIRONMENTAL FACTORS AFFECTING THE DISTRIBUTION AND ABUNDANCE OF CHIRONOMIDAE (DIPTERA) IN TWO SEWAGE OXIDATION LAKES

By

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The distribution and abundance of larval Chironomidae were investigated in the first and last lakes of a 4-lake sewage oxidation system. Species composition and adult seasonal occurrence were also examined.

A stratified random sampling program was employed to quantitatively sample larvae in Lakes 1 and 4. Each lake was stratified by depth, distance from the influent site, and wind. Samples were taken during the summer and early fall of 1976 and during late spring and early summer 1977.

Chironominae was the dominant subfamily in both lakes initially, the Orthocladiinae gradually becoming dominant in Lake 4 by early summer 1977. The patterns of larval distribution and abundance in Lakes 1 and 4 indicate that the Chironomidae actively responded to differences in oxygen and nutrient availability in the sediments by migrating to areas offering favorable conditions. Oxygen appeared to be the limiting factor in Lake 1, nitrogen and phosphorus in Lake 4.

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## INTRODUCTION

The Chironomidae are often the dominant macroinvertebrate taxon occurring in sewage oxidation ecosystems. Kimerle and Enns (1968) surveyed 18 waste stabilization lagoons in Missouri and found that more than 94% of the insects in each lagoon were Chironomidae. Average densities of one species, Glyptotendipes barbipes (Staeger), ranged from 40,000 to 269,000 larvae per sq m in some lagoons (Kimerle and Enns 1968, Smith and Enns 1969). Sherberger (1974) reported that the Chironomidae accounted for almost 76% of the insect fauna and more than 41% of its biomass in a Georgian sewage oxidation pond.

The huge midge populations emerging from oxidation systems are a nuisance in warm climates. Mass emergences in Florida and California have restricted man's outdoor activities, discouraging tourism and creating hazardous driving conditions by reducing visibility and making roads slippery (Beck and Beck 1969, Grodhaus 1963). Adults were also reported to disturb feeding livestock (Grodhaus 1975) and induce allergic responses in man (Grodhaus 1963).

The abundance of Chironomidae in sewage oxidation systems is due to their ability to tolerate low oxygen concentrations and the byproducts of sewage decomposition (Sturgess and Goulding 1968). Chironomid larval distribution, largely determined by "planktonic" first-instar dispersal and migrating older larvae (Davies 1976a, 1976b), is influenced by many factors. In sewage oxidation ecosystems they are generally most abundant

in the shallow regions close to shore, at depths of  $< 1$  m (Fagan and Enns 1966, Kimerle and Anderson 1971, Kimerle and Enns 1968, Sherberger 1974). Greater species diversity in shallow zones has been related to a greater diversity of microhabitats (Sherberger 1974) and higher densities to thick algal mats (Fagan and Enns 1966, Kellen 1953, Kimerle and Enns 1968). Also, low species diversity and small populations at depths greater than 1 m may reflect the reduced availability of dissolved oxygen farther from the periphery of stabilization lagoons (Sturgess and Goulding 1969).

The nature of the substrate has also been linked to larval chironomid distribution and species composition. Ali and Mulla (1976) demonstrated that Chironomus spp. preferred substrates of fine sand or filamentous algae to concrete, detrital, and coarse-particle substrates; however, species of Cricotopus proliferated in detritus. Mackey (1976) found the presence of Chironomidae to be in direct proportion to the organic content of the sediments. In high nutrient ponds, the shift from benthic filamentous algae and phytoplankton to higher aquatic plants was paralleled by a shift from large, benthic, algal consumers to smaller, plant-browsing forms (Hall et al. 1970).

The objective of this study was to determine the distribution and abundance of larval Chironomidae in a 4-lake sewage oxidation system, and to examine some of the major factors that may influence their distribution. Species composition and adult seasonal occurrence were also examined.

## MATERIALS AND METHODS

### Study Area

Research was conducted at the Water Quality Management Project (WQMP) study site, located 5 mi south of the Michigan State University (MSU) campus. The WQMP is a multi-faceted system for the tertiary treatment of domestic wastewater. In addition to the series of 4 sewage oxidation lakes, it includes 3 marshes and a tract of land partially equipped for spray irrigation. Wastewater, secondarily treated except for phosphorus, is delivered to the WQMP daily except during the winter. High concentrations of phosphorus and nitrogen remaining in the water are removed and concentrated by photosynthetic and bacterial activity in the lakes.

The WQMP lake system has an area of approximately 16 ha. Each lake has an average depth of 1.83 m and a maximum depth of 2.44 m. These depths were designed to maintain the bottom within the zone of effective light penetration. At present, the hydraulic loading rate is not at the theoretical maximum, 2 million gal per day. It ranged from a high of 400,000 gal per day in 1976 to a low of 40,000 gal per day in 1977. The waste treatment plant effluent flows to Lake 1 and from there through Lakes 2, 3, and 4 in series.

Although the substrate of the lakes was originally clay, organic material has accumulated on the bottoms. The areas of build-up are a function of wind direction and velocity, distance from the influent site,

and organic settling rate (Young and Ball 1978).

Lakes 1 and 4 were selected for larval sampling because of their widely contrasting water quality (Table 1). Phytoplankton and periphytic algae were the predominant primary producers in Lake 1, whereas aquatic macrophytes were predominant in Lake 4 (Bahr et al. 1976). Algae and macrophytes were harvested from each lake during the summers of 1976 and 1977 by WQMP staff.

### Field Studies

#### A. Larval sampling

For the purpose of larval sampling, each lake was stratified by depth, distance from the influent site, and wind, predominantly from the south-southwest during the sampling periods (Figure 1). The depth divisions were as follows: depth I, 0-60 cm; depth II, 61-150 cm; depth III, 151-244 cm. Lakes 1 and 4 were partitioned into 3 and 5 sections, respectively.

Within strata, sampling units were randomly distributed. Numbers were assigned to each box in a grid covering a map of the lake surface. The boxes to be sampled were chosen from a table of random numbers. On the lake, the sampling sites were found by navigating from stakes placed at the points sectioning strata along the lake's perimeter.

Sampling units were optimally allocated among strata as outlined below (Elliott 1971). Preliminary samples were taken from Lakes 1 and 4 in June 1976. The units comprising these samples were allocated among strata in proportion to each stratum's area. The preliminary results were used to calculate the standard deviation for each stratum, and in the subsequent sample, units were allocated in proportion to the respective standard deviations (Elliott 1971). All subsequent samples were

Table 1. The range of average concentrations of selected water quality parameters in Lakes 1 and 4 in the summers of 1976 and 1977.

Parameter, mg/l <sup>1</sup>	Lake 1	Lake 4
Total alkalinity	173.67-201.41	69.48-135.26
Nitrate	3.18- 5.27	0.01- 0.41
Total phosphorus	2.99- 3.29	0.03- 0.36

<sup>1</sup> Parameters were monitored daily by the Institute of Water Research.

Figure 1. Lakes 1 and 4 of the WQMP sewage oxidation lake system.

Sections within the lakes are designated by arabic numerals. Depth contours for the three depth strata are also illustrated by roman numerals as follows:

I, 0-60 cm; II, 61-150 cm; III, 151-244 cm.

optimally allocated in this way (Kimerle 1968).

The number of sampling units taken per sample (Table 2) was calculated with the following formula:

$$n = s^2 / (D^2 \cdot \bar{x}^2),$$

where s is the standard deviation, x is the arithmetic mean, and D is the desired level of precision (Elliott 1971). D was set at a level yielding an error of 15-20% of the mean in all but two cases (25-30%).

A hand-operated, plexiglass core sampler was used to sample benthic Chironomidae. The core of the sampler had an inner diameter of 9.21 cm and an area of 0.0067 sq m. Since 95% of the larvae inhabit the first few centimeters of the substrate (Oliver 1971), only the upper 8 to 10 cm retrieved by the corer were retained for analysis.

Samples were taken every other week from Lakes 1 and 4 during the summer and early fall of 1976. In 1977, the two lakes were sampled once each in May and June.

#### B. Adult sampling

Floating pyramidal emergence traps were utilized to capture adult Chironomidae from all 4 lakes in 1976 and 1977. The trap covered 0.0484 sq m of the water surface. The sides were of white netting with a mesh opening of 240  $\mu$ m. Insects were collected in a container at the top of the trap, removed, and then preserved in 95% ethanol. Traps were placed above depths of 100, 180, and 240 cm, and adults were sampled biweekly from May through September in 1976 and 1977.

Table 2. The number of sampling units taken per sample from Lakes 1 and 4.

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	<u>Date</u>					
	<u>1976</u>			<u>1977</u>		
	<u>July</u>		<u>Aug</u>		<u>Sep</u>	
Lake 1	32	35	42	49	46	
Lake 4	42	51	52	54	45	

---



## Laboratory Studies

### A. Larvae

The larvae in each sampling unit were separated from the substrate by sucrose flotation (Anderson 1959, Charles et al. 1974, Cooper 1965). The sucrose solution containing the larvae was poured through a 250  $\mu\text{m}$  mesh sieve. Larvae were identified to subfamily and stored in 80% ethanol.

The factorial analysis of variance for unbalanced data was performed to determine whether there were significant differences in subfamily distribution with respect to depth or section. Counts per sampling unit were log transformed (Elliott 1971). The simple contrast test was used to determine the source of differences revealed by the ANOVA.

Green's dispersion coefficient (Elliott 1971) was calculated for the Chironomidae on each sampling date. This coefficient is an appropriate index for the comparison of contagiously distributed organisms such as the Chironomidae, because it is unaffected by changes in the number of sampling units, the value of the sample mean, or the sum of all the individuals in the sample (0 = random dispersion, 1 = maximum contagion).

The distribution of biomass among several weight categories was established for each subfamily with the aid of a body length-weight function. Freshly freeze-killed larvae (Pritchard 1976) were sorted into 0.5 mm body length classes, oven-dried at 60<sup>0</sup> C for 48 hr (Dermott and Paterson 1974), and weighed on an electrobalance. The weights were then regressed on the lengths, which ranged from 3.5-4.0 to 22.5-23.0 mm.

Since large and small species were considered together and many first and second instars may have been lacking, a polynomial regression yielded the only acceptable predictive model. The following relationship

yielded the best fit, having an F statistic significant at  $< 0.0005$  and an  $r^2$  of .76:

$$\text{weight} = a_1 \cdot \text{length}^2 + a_2 \cdot \text{length}^3$$

(cf, Mackey 1977).

#### B. Adults

Adult Chironomidae were mounted on slides using the procedure described by Saether (1969) and then identified to species. Identifications were made by D. W. Webb of the Illinois Natural History Survey.

## RESULTS AND DISCUSSION

### Larval Studies

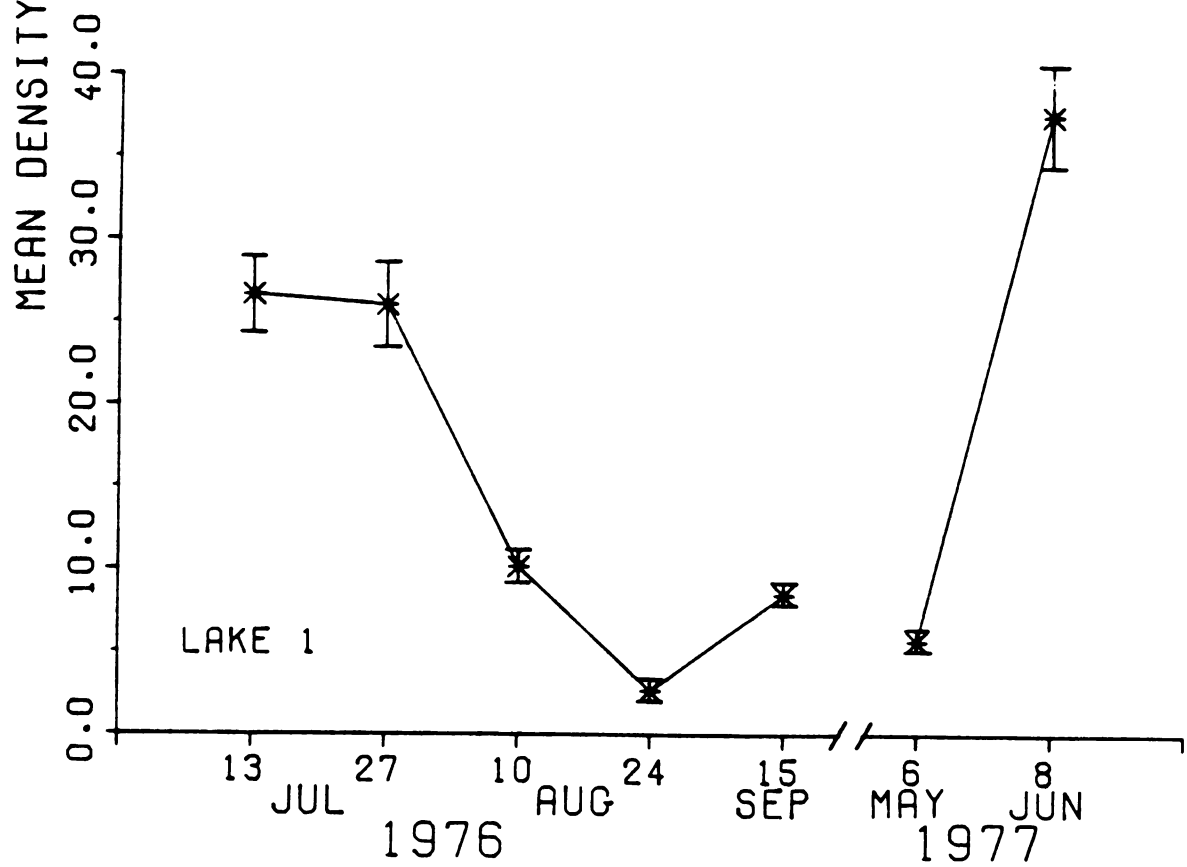
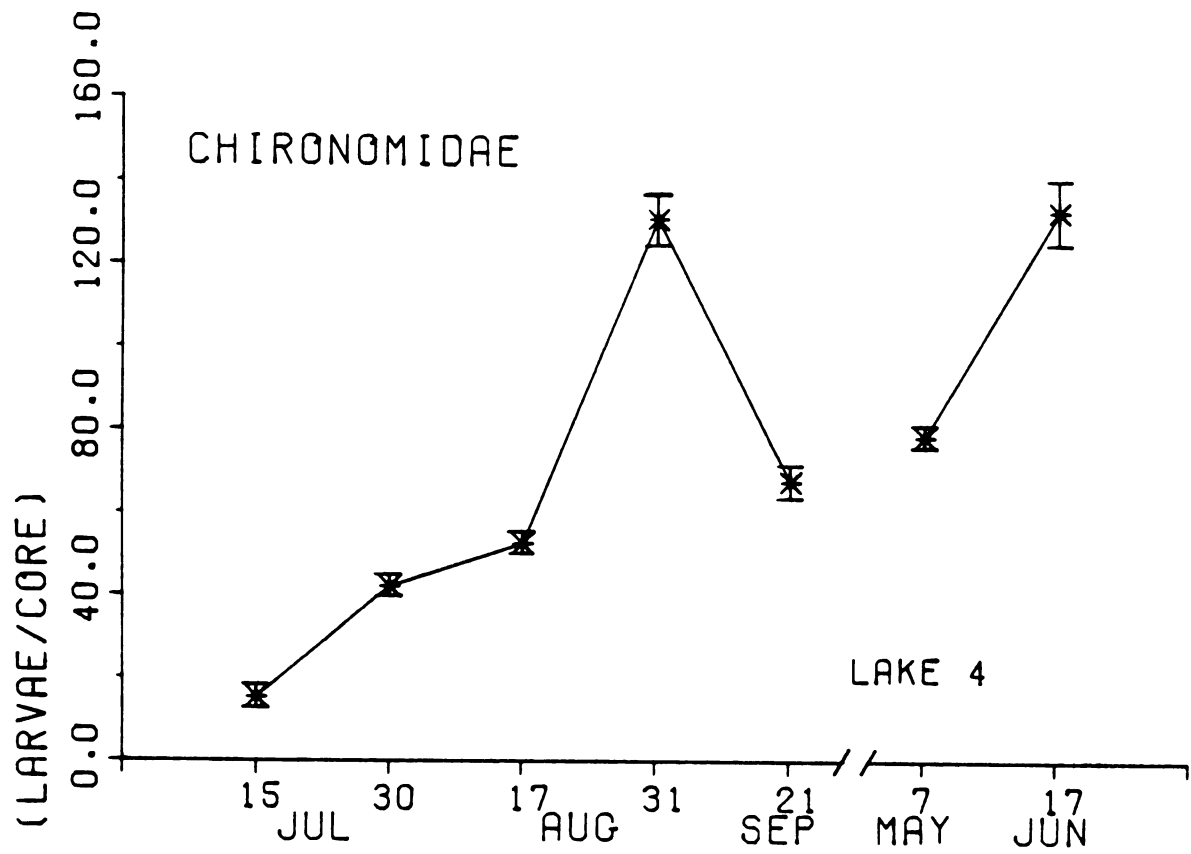
#### A. Lake 1

The mean density of the Chironomidae in Lake 1 reached a plateau in July, then gradually dropped to a low in late August (Figure 2). The mid-September population level appears to have been maintained over the winter. In spring, mean larval density increased rapidly to a peak higher than the previous summer.

Chironominae was the dominant subfamily represented in benthic samples collected from Lake 1, accounting for approximately 70% of the entire chironomid population. The Chironominae and Tanypodinae together constituted over 90% of the larvae collected on each sampling date; the remainder belonged to the subfamily Orthoclaadiinae.

Oliver (1971) noted that species belonging to the Chironominae and Tanypodinae constitute the majority of the species tolerant of very low concentrations of oxygen. As the summer progressed, the sediments of Lake 1 received an ever increasing load of decaying phytoplankton, in addition to very high concentrations of nutrients, e.g. phosphorus. Anaerobic conditions resulting from active benthic decomposition in and near the sediments were not uncommon (Young and Ball 1978) and may have been largely responsible for the dominance of Chironominae and Tanypodinae in Lake 1.

Figure 2. Density fluctuations of the chironomid populations in Lakes 1 and 4. Standard errors about each weighted mean are shown.



ANOVA showed that there were significant differences in the distribution of both Chironominae and Tanypodinae with respect to depth (Figures 3 and 4). Use of the simple contrast test revealed that the Chironominae at depths I and II occurred at comparable densities in both years, but at depth III densities were significantly to very highly significantly different from those at the other depths on most sampling dates. At shallower depths, the Chironominae occurred at the same density through August, then increased markedly in September to a level also found in the spring of 1977. At depth III, density behaved differently, decreasing at a relatively constant rate through August, then increasing in September, though less rapidly than at depths I and II. Also unlike the Chironominae in shallower regions, those at depth III increased abruptly in density in June 1977.

Oxygen availability very likely influenced the distribution of Chironominae in Lake 1. As mentioned earlier, eutrophic conditions in Lake 1 caused benthic oxygen supplies to be exhausted in the summer. However, wind-induced water currents, which scoured the substrate in shallow regions with enough force to resuspend precipitated seston (Young and Ball 1978), probably created more tolerable conditions at shallow depths by frequently re-introducing oxygen. Such mixing also could have occurred in deep areas, but infrequently, because currents at depth III are much less forceful (Young and Ball 1978). Frequent, periodic depletions of oxygen at the substrate-water interface at depth III appears to have prompted early instars to migrate preferentially to shallower, oxygenated areas in August 1976. In July, 20-30% of the Chironominae at all depths were in the lightest weight categories. However, in August the Chironominae in these categories at depth III

Figure 3. Density fluctuations of the Chironominae in Lake 1 at different depths. Standard deviations about each mean are shown.

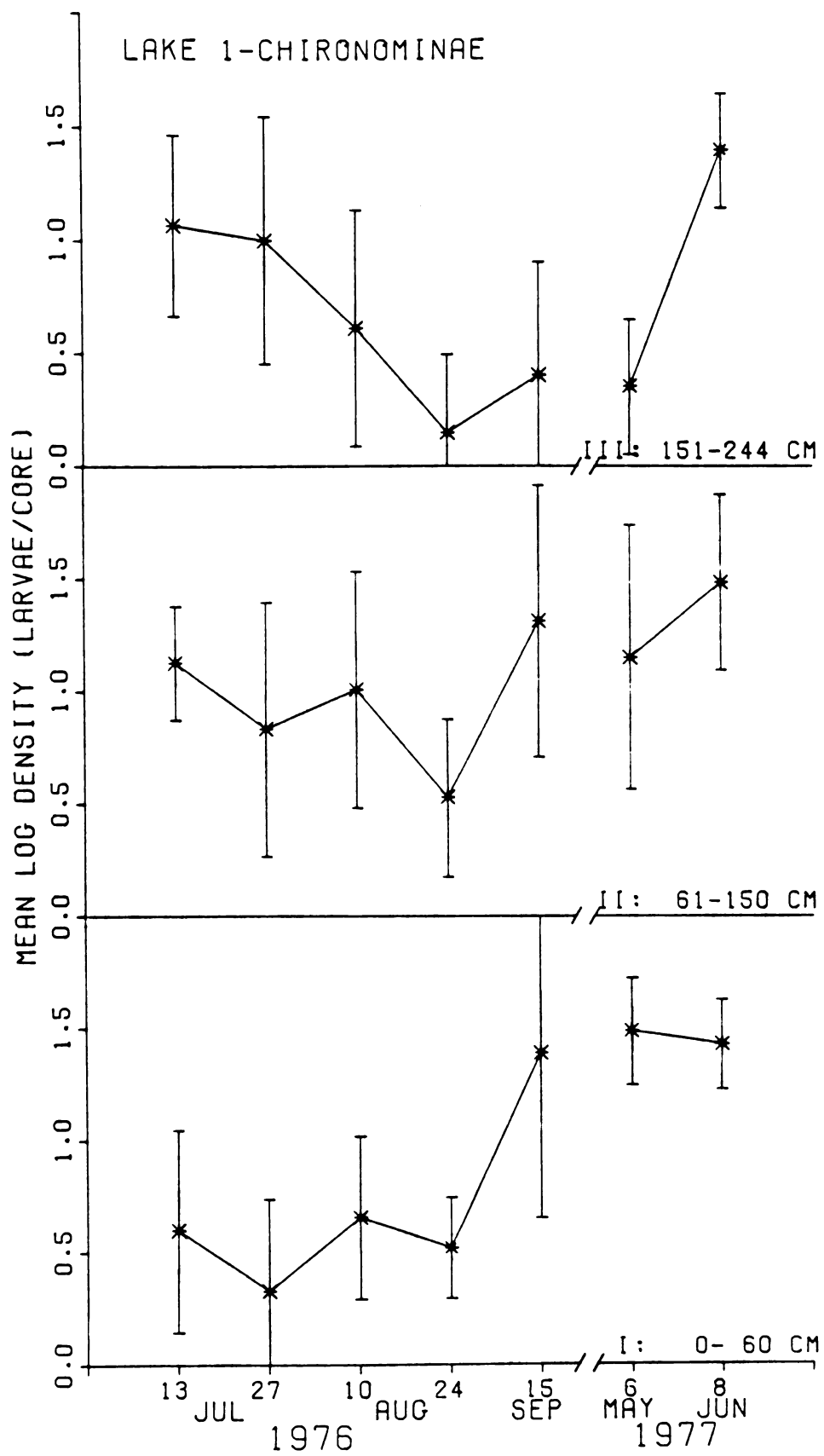
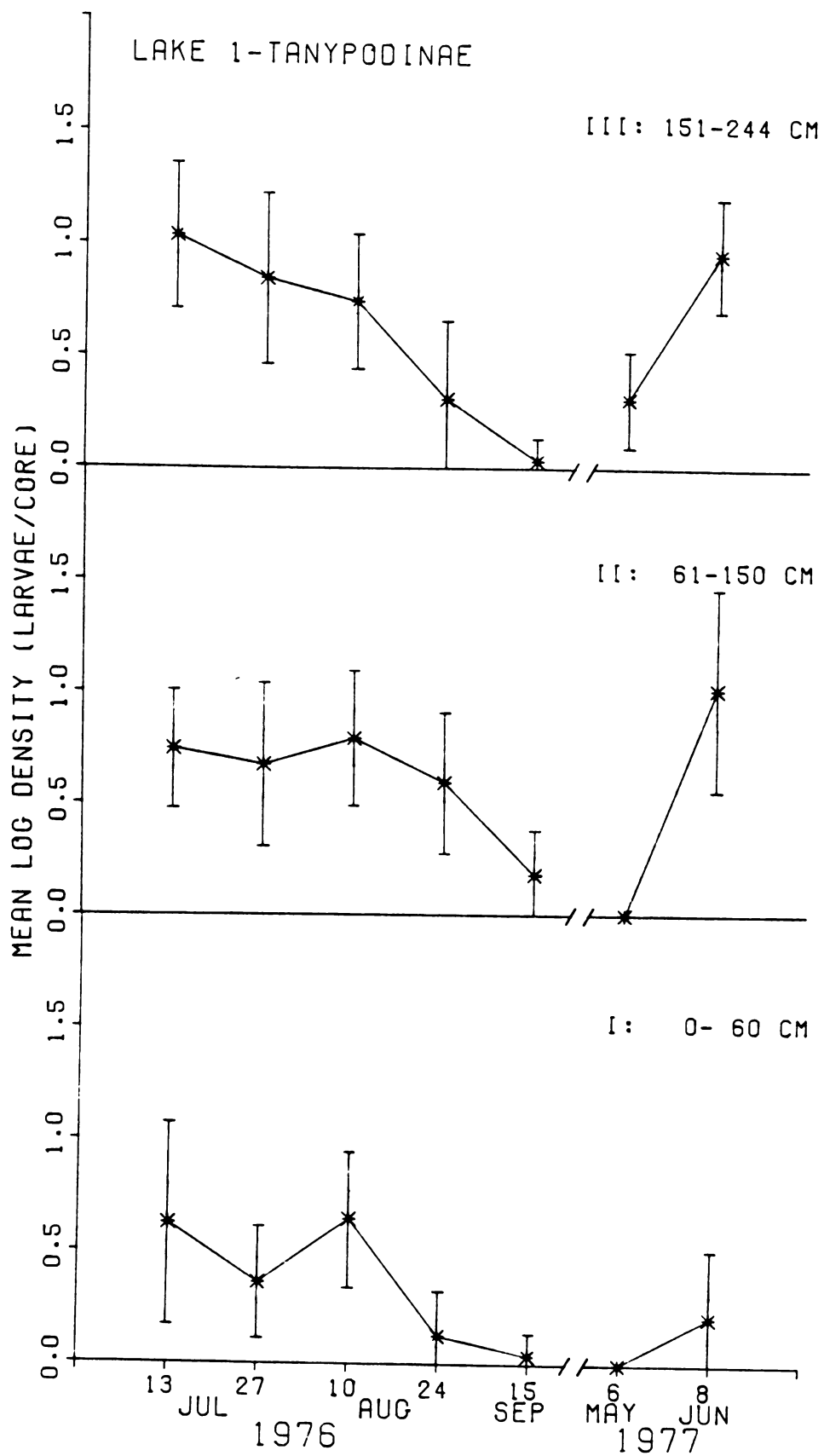




Figure 4. Density fluctuations of the Tanypodinae in Lake 1 at different depths. Standard deviations about each mean are shown.



decreased to 0-10%. The corresponding decline in density at depth III, and its stability at depths I and II (Figure 3), indicate that most of the larvae hatching from eggs laid by new adults migrated to the shallower depths (adult Chironomidae emerged continuously from Lake 1 in summer 1976). Several authors have recorded shifts in maximum chironomid density from profundal to littoral regions in lakes during the summer (Davies 1976a, Eggleton 1931, 1934) and all instars are known to migrate from deteriorating environments to areas offering more favorable conditions (Davies 1976a).

Widespread adult emergence in late August 1976 and May 1977 seems to have been followed by extensive re-colonization by Chironominae in September 1976 and June 1977, respectively. On 24 August and 6 May, there was a shift of biomass towards the heaviest weight categories at all depths in Lake 1 (e.g., Figure 5). On the same dates, there was a reduction in the number of larvae found per sampling unit (Figure 6), indicating that the size of the population clusters was shrinking as adults emerged. Subsequently, there was an increase in larval chironomine density at all depths in September, and at depths II and III in June (Figure 3). The simultaneous increases in the proportion of larvae in the lightest weight categories and in the number of larvae found per sampling unit suggest that the density increases resulted from colonizing young Chironominae. A substantial decrease in the value of Green's coefficient in September, maintained through spring 1977, also supports this hypothesis, since a more random dispersion would be expected to accompany a population expanded by early instars.

The Tanypodinae in Lake 1 responded as the Chironominae had in early summer 1976 (Figure 4). However, the Tanypodinae then showed a

Figure 5. The percent frequency of Chironominae at depth III, Lake 1 in several weight classes, on each sampling date.

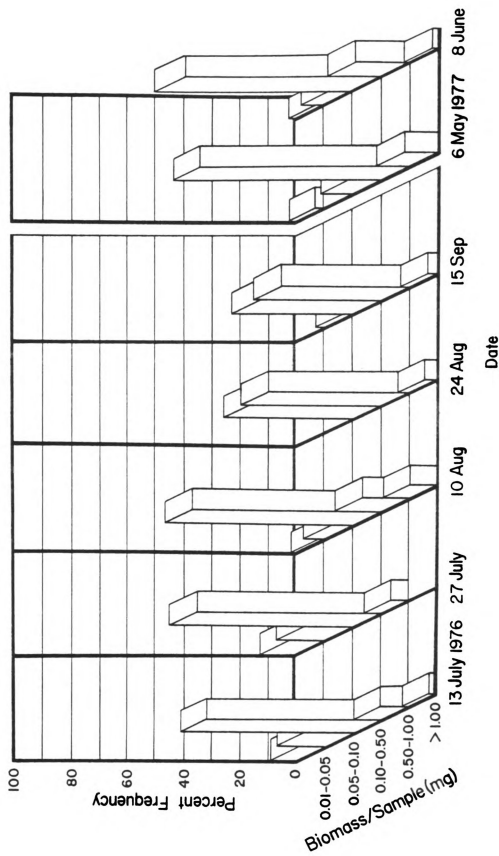
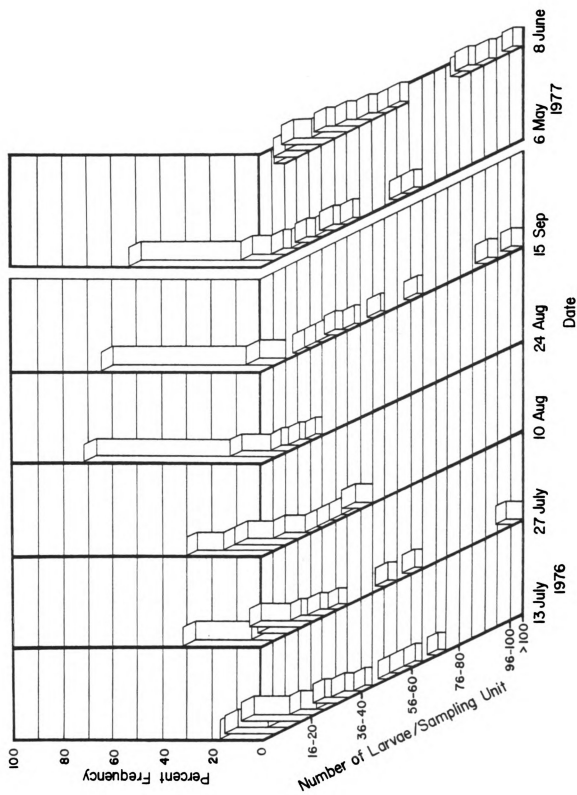


Figure 6. The percent frequency of the numbers of Chironomidae per sampling unit in Lake 1, on each sampling date. The number of larvae per sq m can be obtained by multiplying the number of larvae per sampling unit by a factor of 150.



gradual loss in density at all depths which was not reversed until June 1977. It is difficult to explain the changing distribution and abundance of free-swimming tanypodine predators without knowing the status of their prey populations. The low proportion of larvae in the lightest weight category during the early summer went to zero at all depths but II in late August 1976, indicating that Tanypodinae were emerging in greater numbers than were re-colonizing during 1976. The Tanypodinae biomass shifted in mid-August, similar to that of the Chironominae in late August. Depth II appears to have been the only one subsequently colonized by early instars, as indicated by: (1) the presence of larvae in the lowest weight category, and (2) the significantly higher density in September (Figure 4).

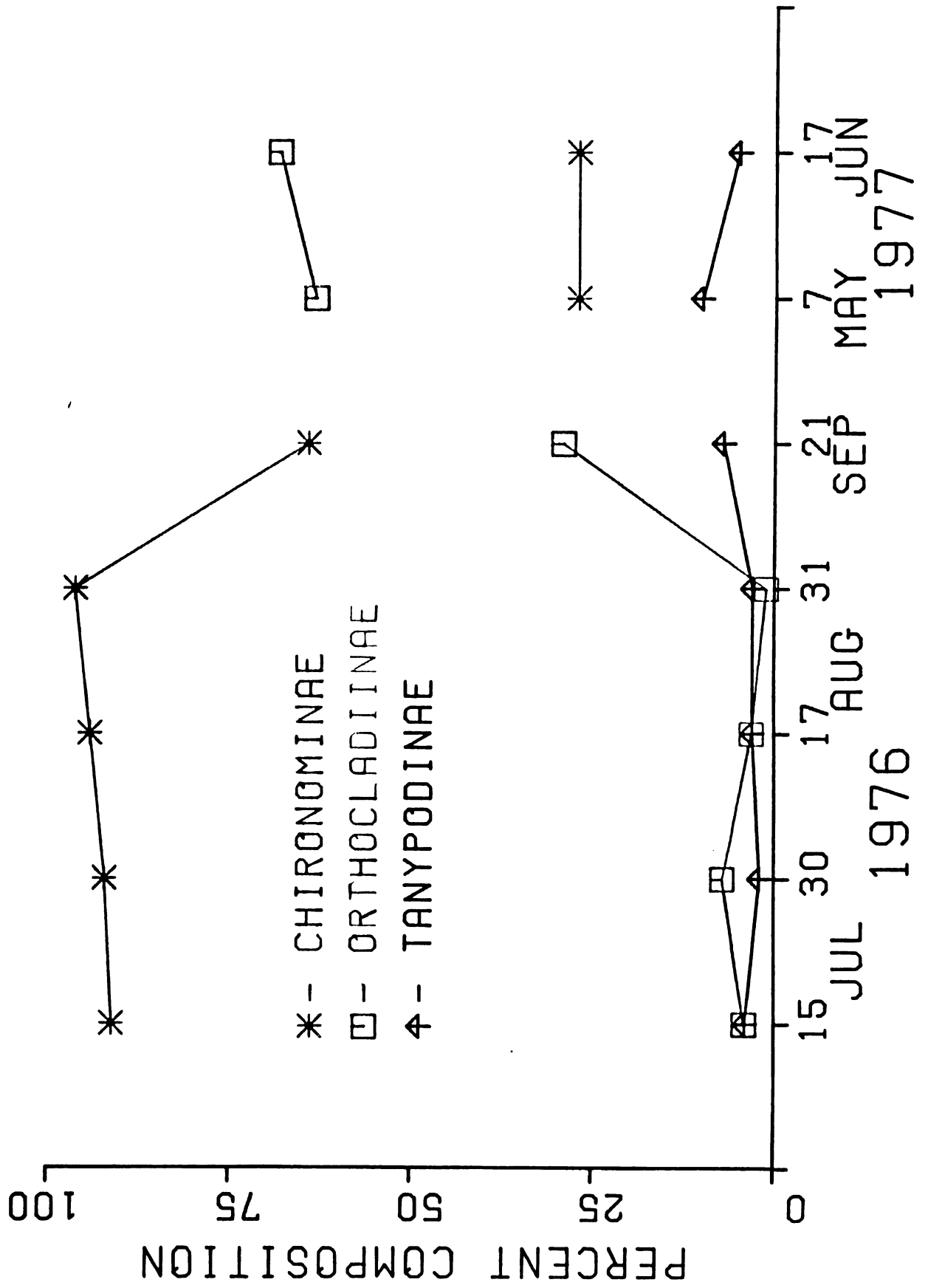
#### B. Lake 4

The mean density of Chironomidae in Lake 4 increased during the summer of 1976, then decreased in the fall (Figure 2). The following spring, population density reached the peak density of the previous summer by mid-June.

The composition of the chironomid community in Lake 4 varied more than in Lake 1, the Chironominae and Orthocladiinae exchanging pre-dominance in fall 1976 (Figure 7). The Chironominae accounted for over 90% of all larvae through August, and for 90% or more of the larvae in each of the lake's 5 sections. In September, the Orthocladiinae represented a greater proportion of the total numbers, averaging 29% over the entire lake and ranging from 16% in section 1 to 36% in section 2. In 1977, the Orthocladiinae increased in numbers and replaced the Chironominae as the dominant subfamily in Lake 4. They constituted over 60% of the



Figure 7. The composition of Chironomidae in Lake 4, by subfamily.



Chironomidae in the lake on each sampling date during 1977 and represented the majority of the midges in each section.

The predominance attained by the Orthoclaadiinae is not readily accounted for. Two of the common orthocladine genera captured in emergence traps on Lake 4, Cricotopus and Psectrocladius (see Table 4), are consumers of higher aquatic plants (Gaevskaya 1969). It is possible that these small orthocladine species were better suited to the dense aquatic macrophytes in Lake 4 than the larger chironomine species. As noted earlier, Hall et al. (1970) found small plant-browsing species to have predominated in environments characterized by higher aquatic plants.

ANOVA showed no consistent pattern of chironomid distribution with respect to depth or section in Lake 4. However, in some instances there were significant to very highly significant differences in the densities of the Chironominae at different depths. The differences were between depths I and III and depths II and III, not between I and II (Table 3), a phenomenon in accord with the growth of the macrophytes. At shallow depths, dense macrophyte beds grew to the water's surface early in the season, creating a sheltered, nutrient-rich environment (Young and Ball 1978). Higher densities of Chironominae occurred at depths I and II early in summer of 1976 and spring 1977 probably as a result.

The twofold increase in the density of the Chironominae at depth III in late August seems to have been related to changes resulting from human interference. In mid-August, the macrophytes across Lake 4 were cut to within 1 ft of the bottom. The harvesting disturbed the water, resuspending organic detritus at all depths. Young and Ball (1978) found seston to be sedimenting at a rate 4 times faster at the end of August than recorded at the beginning, and at the highest rate at depths 1.5 m

Table 3. Density (mean log number/core) of Chironominae in Lake 4 at different depths, on selected sampling dates  $\pm$  SD. Significance as determined by ANOVA included.

Sampling date	Depth I	Depth II	Depth III	Significance
15 JUL 1976	1.42 $\pm$ 0.31	1.31 $\pm$ 0.47	0.90 $\pm$ 0.53	0.0007
31 AUG 1976	1.37 $\pm$ 0.58	1.64 $\pm$ 0.47	1.99 $\pm$ 0.44	0.0011
20 JUN 1977	<sup>1</sup>		1.16 $\pm$ 0.51	0.0334

<sup>1</sup> This depth was not sampled on this date because the water level in the lake had been lowered.

and greater. The evidence indicates that the abrupt density increase resulted from the preference of colonizing young Chironominae for the enriched environment at depth III. The distribution of chironomine biomass in Lake 4 shows that larvae generally shifted to the heaviest weight classes in mid-August. Subsequently, in late August, the coefficient of dispersion dropped to one-third of the value prevailing in previous samples. Apparently then, adults emerged in large numbers from the lake in mid-August, and the larvae hatching from the eggs they laid migrated mostly to depth III. Diffuse macrophytic photosynthesis, in conjunction with very low nutrient loading, produced aerobic conditions at all depths in Lake 4 (Young and Ball 1978).

Larval chironomid biomass in Lake 4 shifted towards the heaviest weight categories in September 1976 and May 1977, once again indicating mass emergence at those times. The overall decrease in larval density also suggests that Chironomidae left the system in September (Figure 2). There was a fourfold decrease in Green's coefficient in May, likely due to colonization by more randomly distributed young larvae. The biomass distribution of Chironominae in sections 3 and 5 exhibited a bimodal peak among the heaviest weight classes during July and late August (Figure 8).

#### Adult Studies

A total of 24 chironomid species comprising 16 genera were collected from the lake system of WQMP. The species collected and their period of emergence in the lakes are listed in Table 4.

Most species emerged continuously during the season, including those most common in Lakes 1 and 4. Several species emerged in one distinct

Figure 8. The percent frequency of Chironominae in section 5, Lake 4 in several weight classes, on each sampling date.

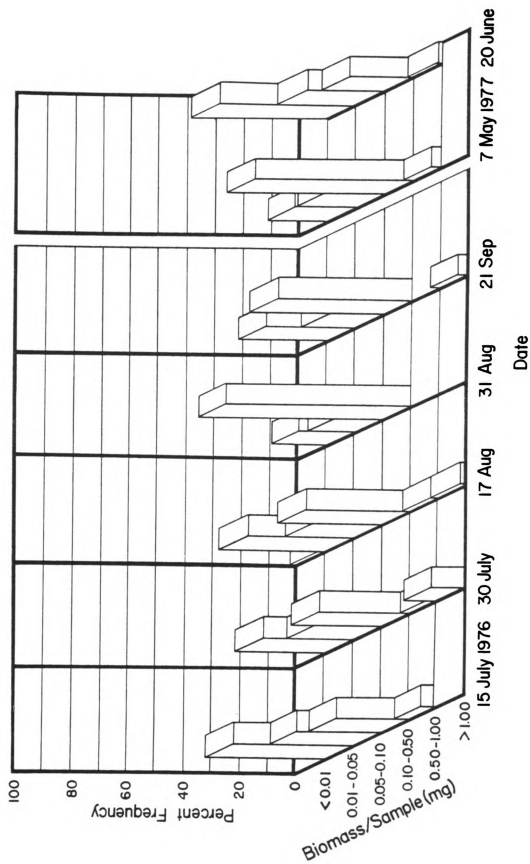


Table 4. Adult emergence patterns of Chironomidae in the WQMP lakes in 1977.

	MAY	JUN	JUL	AUG	SEP
<u>Species</u>					
CHIRONOMINAE					
<u>Chironomus attenuatus</u> (Johannsen)			---		
<u>C. tentans</u> Fab. *†	-----				
<u>Cryptochironomus fulvus</u> Johannsen				---	
<u>C. psitticinus</u> Meigen			---		
<u>C. varus</u> Goetghebuer				-----	
<u>Dicrotendipes nervosus</u> Staeger †	-----				
<u>Glyptotendipes lobiferus</u> (Say) *†	-----				
<u>Parachironomus tenuicaudatus</u> (Malloch) *†	-----				
<u>Polypedilum gomphus</u> Townes				---	
<u>P. illinoense</u> (Malloch)	-----			---	
<u>Pseudochironomus richardsoni</u> Malloch †	-----				
<u>Tanytarsus</u> spp. *†	-----				
ORTHOCLADIINAE					
<u>Corynoneura scutellata</u> Winnertz *†	-----				
<u>Cricotopus bicinctus</u> Meigen	-----				
<u>C. flavibasis</u> Malloch *†	-----				
<u>C. trifasciatus</u> (Panzer) *†	-----				
<u>Psectrocladius vernalis</u> (Malloch) †	-----				
TANYPODINAE					
<u>Ablabesmyia aspera</u> (Roback) †	-----				
<u>A. cinctipes</u> (Johannsen)	-----				
<u>Clinotanypus plannus</u> Roback			---		
<u>Procladius bellus</u> (Loew)		-----			
<u>P. freemani</u> Sublette *†	-----				
<u>Labrundinia pilosella</u> (Loew) †	-----				
<u>Labrundinia</u> sp. †	-----				

\* Occurred in more than 10 of 80 samples collected from Lake 1 in 1977.

† Occurred in more than 10 of 90 samples collected from Lake 4 in 1977.



period, e.g., Chironomus attenuatus (Johannsen). One species, Polypedilum illinoense (Malloch), emerged in two distinct periods, first in May and June, then in August.

## SUMMARY AND CONCLUSIONS

As benthic organisms, Chironomidae are subject to the environmental conditions prevailing at the substrate-water interface. The patterns of larval distribution and abundance in Lakes 1 and 4 indicate that the Chironomidae actively responded to variations in oxygen and nutrient availability in the sediments.

Oxygenation of shallow depths occurred in Lake 1 because phytoplankton and periphytic algae did not block wind-induced water currents; oxygen was depleted at depth III due to the accumulation of decaying phytoplankton and seston. As a result, Chironominae apparently migrated to shallow areas in response to the higher oxygen concentrations prevailing there.

Aerobic conditions characterized all depths in Lake 4 as a result of the predominance of aquatic macrophytes. Nutrients were retained at depths I and II because of macrophyte growth in early spring and summer. The absence of macrophytes across the lake in mid-August 1976 was partially responsible for the substantial deposition of seston at depth III. In Lake 4, nitrogen and phosphorus availability, not oxygen, were apparently the limiting factors, prompting larvae to migrate preferentially to depths I and II early in the summer, to depth III later.

A sewage oxidation system is a continuum from highly enriched to less enriched waters. Therefore, environmental factors other than enrichment must be manipulated if Chironomidae within such systems are to

be kept at acceptable levels. The results of this study indicated that depth, having a significant impact on the distribution and abundance of Chironomidae, is a design variable than can be managed to reduce chironomid population size. If the depth of the water in highly enriched lakes completely exceeds the trophogenic layer, a hypolimnion devoid of oxygen will develop and will effectively limit the abundance of Chironomidae. Unfortunately, increasing the water depth will decrease the efficiency of nutrient reclamation. This situation might be remedied, however, by adding to the number of lakes comprising the oxidation system.

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