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An in situ assessment of the impact of chlorinated wastewater on the macroinvertebrates inhabiting the upper hyporheic zone /

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AN IN SITU ASSESSMENT OF THE IMPACT OF CHLORINATED WASTEWATER ON THE MACROINVERTEBRATES INHABITING THE UPPER HYPORHEIC ZONE

A Thesis Presented

by

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To my

Wife

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ABSTRACT

AN IN SITU ASSESSMENT OF THE IMPACT OF CHLORINATED WASTEWATER ON THE MACROINVERTEBRATES INHABITING THE UPPER HYPORHEIC ZONE

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The macroinvertebrates inhabiting the sediment in Lampson Brook, Belchertown, were sampled in order to assess the impact of chlorinated wastewater, as compared to non-chlorinated wastewater, on this community. Samples were collected four times, once at the end of the chlorination period and three times following the autumnal termination of chlorination. Samples were collected from four stations, one above the sewage outfall (control), and three below the outfall.

Historically, Total Residual Chlorine (TRC) ranges from 0.82 ppm at station 2 to below detection at station 4. Sediment structure remained similar at all stations through out this investigation.

The greatest impact occurred at station 4, 3700 meters downstream from the sewage outfall. This impact is evident by the decrease in abundance of Total Organisms and Chironomidae (Diptera), diversity, taxa richness, and changes in community structure. Toxicity might occur through three modes of action; direct toxicity of TRC, indirect toxicity through the formation of

TOX, and indirect toxicity through the disruption of the food chain. One or all avenues of impact may be occurring.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1

INTRODUCTION

1.1 History of Chlorination

As early as 1879 chlorine was used for wastewater disinfection (White, 1978). Over the intervening decades it has become a common disinfectant used for the benefit of the community (Chamberlin, 1948; Kittrell and Furfari, 1963; Chambers, 1971). Its applications include the "purification" of potable water as well as the disinfection of wastewater. Though ozonation remains a viable alternative, chlorinations' high benefit per dollar makes it the process of choice for wastewater treatment. The use of 187,00 tons of chlorine for sewage treatment in 1973 is testament to its pervasiveness in the public health domain (Chlorine Institute, 1974). Similar to most states, Massachusetts requires the disinfection of wastewater during the summer months (April to October). However the discharge of a broad spectrum biocide, such as chlorine, is counter to the goal of "...The protection and propagation of fish, shellfish, and wildlife..." promulgated by the 1972 and 1977 Amendments (the Clean Water Act (CWA)) to the Federal Water Pollution Control Act (FWPCA) of 1948 (Rand and Petrocelli, 1985 p.591). In this light, the Commonwealth may well be a major violator of these statutes.

1.2 Chlorine Chemistry

An understanding of chlorines chemical behavior in the environment is fundamental for an understanding of its toxicity. The

two principle factors affecting the fate of chlorine in freshwater are pH and the presence of reactive substances, most notable of which are ammonia, organic nitrogen, organic compounds, and reduced inorganic compounds. In the absence of reactive compounds, at ecologically relevant pH's (i.e. pH >3), only free chlorine is present. Hypochlorous acid (HOCl) and the hypochlorite ion (OCl^-) are the predominant forms of free chlorine. These two forms are equally abundant at pH 7.5. As the pH decreases the acid (HOCl) increases and the ion (OCl^-) decreases. Clearly, natural and wastewaters are not void of reactive substances. In the presence of ammonia, combined chlorine compounds are formed. Chloramine speciation is pH dependent and is a combination of monochloramines (NH_2Cl), dichloramines ($NHCl_2$) and trichloramines (NCl_3). Although free chlorine is the most reactive and toxic form (Brungs, 1973), all three chloramine compounds are toxic (Larson et al., 1978; Seegert and Brooks, 1979). Total Residual Chlorine (TRC) is the combination of free and combined chlorine. Because the toxicity of free chlorine is of approximately the same order as combined chlorine (Merkens, 1958), the measure of TRC is sufficient for the determination chlorine toxicity (Douderoff and Katz, 1950).

The presence of organic compounds also affects the fate of aqueous chlorine. If, as in municipal wastewater, the organic content is high a variety of halogenated compounds (TOX) are created (Jolley, 1975; Murphy, 1975; Oliver and Visser, 1980; Boyce and Hornig, 1983; Kintslyet al., 1983; Miller and Uden, 1983; Trehey et al., 1986). The end products of chlorination may be entirely different from the original materials (Enslow, 1932). Even ground water and

comparatively clean surface waters provide sufficient precursors for the production of halogenated organics (Bjorseth et al., 1977; Picer et al., 1987a and 1987b). Wang et al. (1987) found humic acid and algae to be critical precursors in the formation of nonvolatile halogenated organic compounds in raw lake water.

1.3 Chlorine Toxicity

Toxicity studies of the individual by-products of wastewater chlorination have been sparse (Trabalka and Burch, 1979). The physiological stress of one common by-product, trichloracetic acid, to the dragonfly nymph Somatachlora cingulata was deduced from the increased consumption of oxygen and excretion of ammonia (Correa et al., 1985). The effective concentrations (10 -100 ug l^{-1}) were comparable to those attainable from the chlorination of municipal wastewater.

The damage to non-target organisms by the chlorination of effluents spawned a concommitant increase in research aimed at elucidating the scope and etiology of chlorine toxicity. The first investigation of the toxicity of chlorinated municipal sewage effluent on downstream aquatic biota centered on fish (Tsai, 1969).

Due to their commercial and political importance fish continued to be the focus of most early chlorine studies (Coventry et al., 1935; Merkens, 1958; Arthur and Eaton, 1971; Tsai, 1971; Arthur, 1972; Heath, 1978; Larson and Schlesinger, 1978; Thomas et al., 1980; Trabalka et al., 1980; Osborne et al., 1981;Brooks and Bartos, 1984). Much of the research was focused on determining the tolerance limits of various species to free and residual chlorine.

Brooks and Seegart (1978) classified fish on a scale from sensitive (72 hr. LC50 of 0.35 to 0.71 mg l^{-1}) to resistant (72 hr. LC50 of 1.15 to 1.50 mg l^{-1}) based on the toxicity of intermittent doses of monochloramines. Within this scale the salmonids were the most sensitive. Rainbow trout, Salmo gairdneri, had a 96-hr LC50 of 0.037 mg l^{-1}. This scale, coupled with variations in test species and effective endpoint, results in an overwhelming number of permutations. For example, if chlorine exposure is intermittent it can be expressed as peak concentrations or mean plateau concentrations (Larson and Schlesinger, 1978). In addition to temperature being influential in intermittent toxicity studies (Stober and Hanson, 1974; Brooks and Seegert, 1977; Larson and Schlesinger, 1978), the inherent toxicity of intermittent discharges is generally greater than continuous discharges (Dickson et al., 1974). Although mortality is a clear endpoint, it is a comparatively blunt measure of toxicity. Behavioral responses, which would affect the actual exposure in nature, are a more sensitive endpoint because they provide a gradient, rather than a quantum, response. Some responses observed in fish, such as lethargic swimming, thrashing, bobbing, and distress at the water surface (Dandy, 1972; Brooks and Seegert, 1977) can impair their ability to avoid obstacles, predators, and the chlorinated plume itself (Basch and Truchan, 1976; Brungs, 1976; Larson et al., 1978). The avoidance response (Sprague and Drury, 1969; Meldrim, 1974; Larson et al., 1978) is often delayed (Tsai and Fava, 1975) and may be contingent upon chlorine species (Fava and Tsai, 1976) as well as fish species and temperature (Cherry et al., 1982). Monochloramine concentrations as

low as 5.7 ug l^{-1} elicited a significant avoidance response by medaka (Oryzias lolipes) (Hidaka and Tatsukawa, 1985).

Finer physiological measures have also been applied to the fathead minnow, Pimephales promelas. These include reduced blood pH, Increased lactate levels, lowered arterial pO_2, elevated, slightly, methemoglobin levels, and damaged gill function which resulted in death due to hypoxia (Bass and Heath, 1977). The oxidation of hemoglobin to methemoglobin, as opposed to gill damage, was the cause of toxicity proferred by Grothe and Eaton (1975). They found a one hour exposure to 1.5 mg l^{-1} monochloramine resulted in a loss of equilibrium. Dandy (1972), working with trout, found disequilibrium to be the point of no return (i.e. Removal of the toxicant no longer resulted in recovery).

All trophic levels of the aquatic ecosystem are sensitive to chlorination. Microbial community structure and function were affected at various levels in field enclosures and laboratory microcosms (Pratt et al., 1988). Osborne (1982) observed an increase in respiration and a decrease in primary productivity of whole epilithic communities when exposed to 0.1 mg l^{-1} TRC. Other impacts of chlorine toxicity on phytoplankton include increased mortality (Betzer and Kott, 1969), and reduced photosynthesis (Brook and Baker, 1972), growth rate (Kott et al., 1966), nitrate and ammonia uptake (Toetz et al., 1977), and species composition (Gudzuhn, 1986). Brooks and Liptak (1979) observed an irreversible impairment of the algal photosynthetic system from the loss of chlorophyl-a and the permanent reduction in carbon uptake due to a 30 minute exposure to 1 mg l^{-1} TRC. The aquatic macrophyte

Myriophyllum spicatum is also subject to chlorine toxicity (Watkins and Hammerschlag, 1984). Exposure to 0.05 mg l^{-1} TRC resulted in a 16% reduction in shoot length and a 30% reduction in total plant dry weight.

Macroinvertebrates also have interspecies variations in susceptibility to chlorinated effluents (Arthur et al., 1975; Turner and Thayer, 1980). Unfortunately, much of the research to date with macroinvertebrates has been laboratory oriented (Arthur and Eaton, 1971; Gregg, 1974; Arthur et al., 1975). These laboratory based toxicity tests have provided median lethal concentrations (LC50) for many organisms. The 48-hr LC50 TRC values of zooplankton range from 0.017 mg l^{-1} for Daphnia magna to 3.2 mg l^{-1} for Aeolosoma headleyi (Clarke et al., 1977; Cairns et al., 1978; Ward and Degraeve, 1978). Arthur et al. (1975) observed reduced survivability of Daphnia from exposure to 10 ug l^{-1} TRC in municipal wastewater.

Arthur et al. (1975) found that, in general, the macroinvertebrates have 7-d TL50 values from 0.21 to > 0.81 mg l^{-1} TRC. Amphipods, for example, had a mean long-term no adverse effect level of 12 ug l^{-1} and a mean long-term lowest adverse effect level of 19 ug l^{-1}. Similar values were found in studies without a wastewater component. 7-d LC50 values as low as 0.01 mg l^{-1} have been observed for some invertebrates (Gregg, 1974). The amphipod Hyalella azteca and the crayfish Orconectes australis had a 96-hr LC50 from 0.65 to 0.83 mg l^{-1} TRC and 1.08 mg l^{-1} TRC, respectively (Clarke et al., 1977). Overall Daphnia magna, with acute LC50's of 2 to 45 ug l^{-1}, were the most sensitive invertebrates (Arthur et al., 1975; Ward and Degraeve, 1980; EPA, 1985). In general

fish and invertebrates demonstrated comparable ranges of sensitivities (EPA, 1985).

Although laboratory studies are reproducible and less complicated (Odum, 1977; Cairns, 1986), they lack a degree of ecological relevance (Livingston, 1979; Carriker et al., 1982; Bascom, 1982). In light of this, field measurements are critical for assessing community level reactions to stress (Connell and Sousa, 1983; Likens, 1985; Cairns, 1986; Perry, 1988). Although some routine rapid bioassesment studies will include chlorinated effluents, detailed field investigations into the effects of chlorinated municipal wastewater on the macroinvertebrate community of the receiving stream have been sparse. Simpson (1980) examined gill damage to net spinning Trichoptera. Moore et al. (1980) documented the mutagenicity of macroinvertebrates below a chlorinated sewage discharge. Lewis (1986) made only a cursory analysis of the macroinvertebrates while Pagel and Langdon's (1981, p.11) research was limited to a preliminary study "...designed to identify possible areas in which problems were occurring." Keefe et al. (1983), on the other hand, performed a detailed investigation but their data were limited because they were unable to adequately control for sediment grain size. Osborne (1985) and Osborne and Davies (1987) did make intersite comparisons of macroinvertebrate communities. In this instance, however, the sewage outfall was just below a thermal discharge. Because temperature exerts a profound effect on chlorine toxicity (Capuzzo, 1977; Heath, 1978; Capuzzo, 1979) interpretation of these findings is tendentious. These studies were limited to a distance of 1.5 KM

downstream from the outfall. Furthermore an ultimate dilution factor of greater than 200 occurred, and, although the hyporheic zone is regarded as a refuge for macroinvertebrates, these studies dealt primarily with the river bed surface.

Brungs (1973) reviewed chlorine toxicity and made interim water quality criteria recommendations for continuous and intermittent chlorine discharges. Continuous TRC concentrations of 0.01 mg l^{-1} and 0.002 mg l^{-1} were believed to be protective of resistant and intolerant organisms, respectively. The most restrictive recommendations for intermittent discharges were for those containing free chlorine. To protect salmonids, Brungs suggested TRC should not exceed 0.01 mg l^{-1} for 30 minutes. Mattice and Zittel (1976) recommended a freshwater chronic toxicity threshold of 0.0015 mg l^{-1} TRC. The 1984 ambient water quality criteria for chlorine (EPA, 1985) do not distinguish between continuous and intermittent discharges. It recommends that a four day average TRC concentration of 0.011 mg l^{-1} not be exceeded more than once every three years.

Recent studies of Lampson Brook in Belchertown Massachusetts, Hampshire County, yielded some surprising results concerning the chlorinated sewage discharge (Chang, 1989; Coler, 1990; Reckhow, P.C.). Analysis of the macroinvertebrate communities that colonize suspended artificial substrates indicated a residual toxicity of chlorinated wastewater well beyond the range of TRC and ammonia persistence. Based on the detection of increased levels of organohalides in the sediments below Belchertown Wastewater Treatment Plant (Reckhow, P.C), it was speculated that

this toxicity may be the result of chlorinated by-products retained in the sediments but not in the water column (Coler, 1990).

1.4 Pollution indicators

Although no organism, or group of organisms, are universal pollution indicators, macroinvertebrates are sensitive to the majority of wastes entering our waterways (Tesmer and Wefring, 1981). Their usefulness is based on their key ecological role (Resh and Rosenberg, 1984), especially with regards to the stability of fish populations, habitat preferences and comparatively long life cycles (EPA, 1973; Pratt and Coler, 1976). The macroinvertebrate community that inhabits a specific aquatic ecosystem is the product of its environment (Tesmer and Wefring, 1981). Consequently, changes in the quality of the environment will result in characteristic changes in community structure (Hynes, 1960).

Within the macroinvertebrate community the family Chironomidae (midges) is often the single most abundant and diverse family (Pinder, 1986). This observation, coupled with their ubiquity, has made them the focus of many pollution monitoring studies. Chironomidae have been used as water quality indicators in lentic (Brinkhurst, 1974; Saether, 1979; Aagaard, 1986) and, more recently, in lotic (Learner et al., 1971; Wilson and Bright, 1973; Morris and Brooker, 1980) environments. Although the species richness and diversity of Chironomidae may not follow that of the macroinvertebrate community as a whole (Rosenberg, 1974; Lenat, 1983), the analysis of this family alone has been shown to be sufficient for differentiating between stations with varying degrees

of pollution (Beck, 1977; Armitage and Blackburn, 1985; Kawai et al., 1989). The community level response of Chironomidae is often characteristic of specific heavy metals (Surber, 1957; Wentsel et al., 1977; Winner et al., 1980; Armitage, 1985). The chironomid response to petroleum pollution has also received attention (Tubb and Dorris, 1965; Rosenberg and Wiens, 1976). It is even possible to use morphological abnormalities to identify the effects of industrial and agricultural wastes (Hamilton and Saether, 1971; Warwick, 1985). However, Pinder (1986) concluded that, in general, the effect of organochlorine compounds on the Chironomid community has not been widely investigated.

Although the hyporheic zone has been regarded as a refuge during severe surface conditions (Ford, 1962), especially for small macroinvertebrates, detailed study of the hyporheos (the fauna of the hyporheic zone (Williams and Hynes, 1974)) is relatively new (Williams, 1984). It is now evident that this zone, along with drift, oviposition, and upstream migration, is a principle source for recolonization of the substrate surface (Williams and Hynes, 1976). In a gravel stream with a meter deep hyporheic zone the upper 10 to 20 centimeters were the most productive. However the depth of the hyporheic zone is dependant on the underlying streambed and may be quite limited (Williams, 1984; Williams and Hynes, 1974). Pore size, which is dependent on the size, shape, and compaction of the substrate, is the controlling factor in the hyporheic zone. The larger spaces near the surface may result in increased habitable area (Williams, 1984). A diverse array of Chironomids inhabit all types of

hyporheic substrates (Tavcar and Mestrov, 1970) and are generally the most common insect (Williams, 1984).

Censusing of hyporheic invertebrates with an emphasis on Chironomidae has not previously been done on streams impacted by chlorinated wastewater. Consequently, it may be that the macroinvertebrate community inhabiting the upper hyporheic zone would yield a more sensitive indication of a sediment borne environmental stress than the population in the water column.

CHAPTER 2

OBJECTIVES

I investigated the effects of chlorinated and non-chlorinated wastewater on the composition of the macroinvertebrate community inhabiting the upper hyporheic zone of Lampson Brook, a second order stream in Belchertown Massachusetts. Specifically I did the following:

1) Conducted a survey of the macroinvertebrates inhabiting the riffle of a stream that received chlorinated and non-chlorinated sewage at different times of the year. I also determined the density, diversity, and composition of these invertebrate communities. I placed particular emphasis on the Chironomidae (Diptera: Chironomidae).

2) Determined the association between the macroinvertebrate community structure and the changes in the treatment of the wastewater effluent.

CHAPTER 3

SITE DESCRIPTION

3.1 Lampson Brook and Belchertown, Massachusetts.

Lampson Brook is a second order stream located in the central western portion of Belchertown, Massachusetts (Hampshire County) in the state reservation.

The swamp adjacent to the intersection of State Street and Hill Road is the headwaters for Lampson Brook. The stream passes by the Belchertown State School and is joined by a small tributary, which originates in the small swamp south-west of the intersection of routes 9 and 202, before crossing George Hannum Street and passing the wastewater treatment plant. The brook enters a swamp approximately 120 meters below the treatment plant outfall, crosses back under George Hannum Street 900 meters below the outfall and parallels Boardman Street until widening into a small pond near Mill Road. After passing two private swimming pools on Mill Road the stream crosses Boardman Street and runs under Rural Road (approximately 3700 meters below the sewage outfall) before entering Forge Pond (U.S.G.S. Topo Map, Belchertown, Massachusetts quadrangle, 1979).

The watershed is characterized by agricultural land, areas of dense mixed hardwood and softwood trees, shallow marsh, unimproved land and clustered, light density residential areas (MacConnell, 1975). The regional climate is characterized by warm humid summers and moderately cold winters with an annual rain fall

of 42 inches (Smith, 1975). The minimum flow occurs during the summer. Recharge occurs during the spring months.

Belchertown has a population of 7,863, an area of 55.4 square miles, and is located in west central Massachusetts (Massachusetts Municipal Profiles, 1990).

3.2 Belchertown Wastewater Treatment Plant.

The Belchertown State School Sewage Treatment Plant is a secondary facility designed to handle a flow of 0.5 mgd. The average daily flow from October 1989 to January 1990 was 0.419 mgd, with a minimum of 0.240 mgd, and a maximum of 1.024 mgd (Belchertown WWTP Monthly Reports, 1989). The BOD and TSS levels are generally less than 10 mg l^{-1}, which are in compliance with the NPDES permit (Bill Trombley, P.C.). However, the levels of nitrogen and phosphorus are not reduced (Chang, 1989). The effluent is chlorinated from April first to October thirty-first for the reduction of pathogens.

3.3 Stream Quality.

Water Quality is basically good above the treatment plant outfall. Although the effluent is of comparatively high quality it does elevate the BOD and nutrient load of Lampson brook. Between the months of April and October the effluent has a variable TRC concentration (Table 12). The levels of phosphorus and ammonia regularly ranged between 1.79 to 2.18 mg l^{-1} and 0.390 to 1.690 mg l^{-1}, respectively. This exceeds the recommended levels of 0.02 mg l^{-1} for phosphorus and 0.05 mg l^{-1} for ammonia (EPA, 1972; EPA, 1980).

3.4 Study stations.

A control and three treatment stations were chosen along Lampson Brook in order to assess the effects of chlorinated wastewater on the macroinvertebrates of the upper hyporheic zone. Sampling Locations:

Station 1 (Control)	80 meters above the sewage outfall.
Station 2	80 meters, one reversing meander, below the outfall
Station 3	850 meters below the outfall.
Station 4	3700 meters below the outfall.

The exact station locations were selected in an attempt to minimize the differences in current and sediment composition among stations.

CHAPTER 4

METHODS AND MATERIALS

4.1 Sampling Schedule.

Samples were taken four times during the fall-winter period, which is the end of the chlorination period. This period was chosen because the macroinvertebrate community is most stable at this time (Lenat, 1987). Furthermore, the density of the hyporheos are at their peak during this period (Williams, 1984). The first sampling period was just prior to the termination of chlorination on 27 October, 1989. A second set of samples were taken on 10 November, 1989, approximately one week after chlorination had ceased. Two weeks after that (24 November, 1989), or approximately three weeks post-chlorination, a third set of samples were taken. Based on the preliminary screening of the first three sets of biological samples a fourth set of samples were taken on 5 January, 1990, 67 days post chlorination.

4.2 Biological Samples.

In order to reduce variability, a stratified random sample design that controls for substrate size (Resh, 1979; Alley and Anderson, 1968; Milbrink, 1974) was undertaken at all stations using a 5.5 cm. diameter Stand-pipe corer (Williams, 1981). Twenty samples per station were collected on the first day of sampling (during chlorination). Eleven samples were collected thereafter at each station on each of the subsequent sampling days. The upper 8 cm. of sediment were collected due to the limited depth of

penetrable substrate at some stations (Reckhow, P.C.) and processing limitations. The cores were immediately transferred to Ziploc plastic bags and maintained at a constant temperature commensurate with stream temperatures until processed.

Due to the extensive amount of time required for sample processing, three samples from each station on each day were live-sorted as a means of screening the biological data. The remainder of the samples were sieved (180 micrometer mesh), preserved with 70% ethanol, stained with Rose Bengal, and stored at four degrees centigrade until sorting was possible. All sediment samples were sorted as follows; 1) Organic matter was separated from mineral matter by sugar flotation (Fast, 1970; Lacky and May, 1971; Pratt, 1977). 2) All floating material was subsequently skimmed with a fine mesh screen and picked, together with the surface of the sugar solution, under 70 power magnification. 3) The macroinvertebrates were stored in 70% ethanol in 7-ml vials until identified. 4) Oligochaetes and Chironomidae larvae were mounted on glass slides for identification. The CMC-10 mounting procedure was performed in accordance with protocol suggested by the Massachusetts Department of Environmental Protection (DEQE, 1989). If a sample had more than twenty Chironomidae, a random sample of twenty was selected for identification. Identifications were made to the lowest feasible level using appropriate keys (Pennak, 1978; Brinkhurst, 1986; Weiderholm, 1983; Oliver and Roussel, 1983; Merritt and Cummins, 1984).

4.3 Chemical Data

The thoroughness and consistency of the chemical data generated on Lampson Brook by our laboratory over the past four years (Chang, 1989; Coler, 1990), along with data from external sources (Reckhow, P.C.), precluded the necessity to implement a full battery of chemical analyses. Consequently, the following analyses were performed a minimum of once to verify the validity of the accumulated base line data: field D.O. levels (YSI model 54a Oxygen Meter), field pH levels (Beckman Expand-mate pH Meter), current velocity (f581 Water current Meter), sediment size characterization, temperature, and ammonia-nitrogen levels. Samples for ammonia analysis were collected in polyethylene bottles and stored at four degrees centigrade until analyzed in accordance with the procedures outlined in Standard Methods (APHA, 1985).

Chlorine data generated at the treatment plant were subsequently inspected to assure that chlorination practices had remained constant. One sediment sample from each station on 27 October, 1989 and 5 January, 1990 was dried at 130 degrees centigrade and classified using nested sieves with mesh opening diameters of 6.3, 2.0, 0.50, and 0.075 mm. As defined in Biological Field and Laboratory Methods (EPA, 1973) the general classification of these size catagories is: >6.3 mm = medium gravel and up, 2.0-6.3 mm = fine gravel, 0.50-2.0 mm = coarse sand, 0.075-0.50 = very fine to medium sand, <0.075 = silt and clay. The percent contribution of each catagory to the total weight was then determined.

4.4 Data Analysis

4.4.1 Statistical Analysis.

Descriptive and inferential statistics were performed on a Macintosh SE using the FASTAT 1.0 (SYSTAT, 1989) statistical package. Diversity, percent community similarity, richness, and total and percent abundance were calculated on the Microsoft Works' spreadsheet application for the Macintosh.

Log transformations have been advocated for the analysis of benthological data (Elliot, 1977). However, a comparison of the date by site interactions resulting from Two-way ANOVA of non-transformed and log(n+1)-transformed abundances of the Total Organism yielded no differences at the $P< 0.05$ level of significance (Non-transformed: $F=5.92$, $df=9$, $P=0.000$; Transformed: $F=7.31$, $df=9$, $P=0.000$). A similar trend occurs for Chironomidae abundance (Non-transformed: $F=3.43$, $df=9$, $P=0.001$; Transformed: $F=4.02$, $df=9$, $P=0.000$). Transformed data may yield more highly significant results (i.e. increased F-values). Since $P<0.05$ is sufficeint for the purposes of this investigation the original scale was used for ease of interpretation and prevention of artificial interactions (Miller, 1986).

A similar comparison was made for the inter-site t-tests. Log(n+1)-transformations of the abundance of the selected taxa (i.e. the genera of Chironomidae) did not change the determination of significance at the $P<0.05$ level. Consequently the original scale was used for all comparisons and all subsequent references to statistical significance is at the $P<0.05$ level only.

4.4.2 Calculation of Diversity

Although the Shannon-Weiner index of diversity, H', is criticized for its lack of biological relevance (Washington, 1984), I used it for intersite comparisons because of its pervasiveness in the literature and its conservative correlation with controlled toxicity tests (Perkins, 1983). As defined by Washington (1984), the Shannon-Weiner formula for population diversity based on a sample is:

$$H' = -\Sigma(n_i/n * \ln * n_i/n)$$

Where: n = The number of individuals in the sample from a population.

n_i = The number of individuals of a species i in the sample from a population.

Due to the relatively small sample size when diversity is calculated for individual cores, the sum of all cores for each station on each day was used. Using this method only a point estimate of the population diversity can be derived.

4.4.3 Calculation of Community Similarity

I used the Percent Community Similarity (Psc) index to compare the macroinvertebrate community structure of the stations to themselves through time, and to each other on each date. Psc is sensitive to shifts in dominant and semi-dominant species and, therefor, is a good measure of structural-functional changes (Brock, 1977). The formula for Psc is:

$$Psc = 100 - 0.5 \sum Abs(a - b)$$

Where a and b are, for a given species, the percent of the total samples A and B which that species represents (Whittaker and Fairbanks, 1958). Psc was calculated using the pooled core samples as the sampling unit.

4.4.4 Calculation of current velocity

Current velocity was determined in accordance with the formula:

$$V= 0.226 * N + 0.007 = M/ sec$$

Where N = Revolutions of propeller per second = (B * 20) / 60

Where B = Beeps per minute, and 1 Beep = 20 Revolutions.

CHAPTER 5
RESULTS

5.1 Physico-Chemical Data

Chemical parameters at the control station were generally in compliance with the standards for the protection of aquatic life and public health recommended by the EPA (1986), with the exception of phosphorous and ammonia, which were slightly elevated (Table A.1).

Stations two through four received enrichment from domestic sewage at all times. This sewage was chlorinated from April to October. The organic burden was reflected in an increase in B.O.D., TRC, ammonia, phosphorous, and nitrate relative to the control (Tables A.1 and A.2).

Station three had lower, although not limiting, D.O. levels during the chlorination period relative to the control (Table A.2). A trace amount of TRC was occasionally detected yielding an average of 0.01 mg l^{-1} ($\pm$ 0.003). Ammonia, although still higher than recommended, was not significantly greater than the control. B.O.D. and nitrate were not greater than the control. Phosphate was higher, although not significantly, during chlorination than during the winter and both periods were greater than the control (Tables A.1 and A.2).

During both the winter and the chlorination period station 4 was comparable to the control station with respect to D.O., B.O.D., nitrate, ammonia, and TRC. Phosphate was elevated during chlorination but not during the winter (Tables A.1 and A.2).

Ammonia levels measured during the chlorination period on 27 October, 1989 were lower, except for station 2, than those measured in 1988 (Table A.2). Stations one, two, three, and four had ammonia levels of 0.04, 0.57, 0.08, 0.02 mg l^{-1}, respectively. D.O. levels on 27 October, 1989 were 10.8, 10.2, 5.5, and 10.8 mg l^{-1} for stations one, two, three, and four respectively. On 5 January, 1990 the values measured at these stations were 12.6, 12.0, 8.2, and 12.2 mg l^{-1}. On 27 October, 1989 the pH was 6.5 for stations one, two and three and 6.0 for station four. On 5 January, 1990 the pH was 6.4, 6.2, 6.2, and 6.6 for stations one, two, three, and four respectively. The temperature at stations one, two, three, and four was 9, 11, 10.5, and 11 on 27 October, 1989 and 2, 2.5, 1, and 1 degrees centigrade on 5 January, 1990, respectively. Current velocities were 0.46, 0.46, 0.23, and 0.31 m sec^{-1} for stations one, two, three, and four respectively on 27 October, 1989 .

The physical characterization of the sediments and the interstation comparisons using percent similarity (as defined by Psc) are provided in tables 1 and 2 respectively. On 27 October, 1989 stations two, three, and four were 80, 81, and 69 percent similar to the control. On 5 January, 1990 the similarities of stations two, three, and four decreased to 74, 79, and 67 with respect to the control. The control, on 5 January, 1990, was 89 percent similar to the control on 27 October, 1989. Station four, on 5 January, 1990, was 90 percent similar to station four on 27 October, 1989. Station four was slightly coarser, as shown by the relatively high percentage of fine gravel and coarse sand (Table 1).

5.2 Biological Data

5.2.1 Total Abundance

5.2.1.1 Total organisms

A Two way ANOVA yielded significant differences ($P < 0.05$) for station, date, and interactions. A plot (Fig. 1) of the date by station means and standard errors identifies the source of the significant interactions (e.g. Station 2 behaves differently across time as compared to the other stations.) Furthermore, the plot demonstrates the reasons for the significant date and station effects (e.g. station 2 is usually greater than station 1 and station 1 does not vary appreciably across time whereas station 4 is characterized by date specific changes.)

Although the control did not differ significantly through time, all control treatment comparisons are made using the appropriate observed control mean from the given date. This avoids both ambiguity in the definition of "control" as well as the possible confounding effects of seasonal variation. All subsequent determinations of statistical significance were performed in a like manner.

During chlorination (27 October, 1989) Total Organisms abundance at station 3 was significantly higher than the control, station 2 was significantly higher than both the control and station 3, and station 4 is significantly lower than the control, as well as stations 2 and 3 (Figs. 1 and 2; Table 4). An examination of the data collected from the first two sampling dates during the post-chlorination period (10 November, 1989 and 24 November, 1989) yielded no significant differences among stations (Fig. 1; Table 4).

On the final sampling date (5 January, 1990), stations 2 and 3 were significantly higher than the control (station 1) and station 4 but not different from each other (Figs. 1 and 2; Table 4). Core densities at station 2 decreased (10 November, 1989 and 24 November, 1989) and then increased while the other stations remained constant or increased. In summary, the four stations were significantly distinct during chlorination but, after exposure to approximately two months of non-chlorinated wastewater they separated into two significantly distinct groups.

5.2.1.2 Chironomidae

The general shape of the plot of Chironomidae mean abundance and S.E. (Fig. 3) was similar to the Total Organisms plot (Fig. 1) with a few notable exceptions. During chlorination, station 3 was not significantly different from the control. Although no stations were significantly different from the control on 10 November, 1989, the means for stations 2 and 4 were the highest and lowest values, respectively. Unlike the total abundance data, station 4 Chironomid populations were significantly lower than those of the control on the second sampling date during recovery (24 November, 1989) (Fig. 3; Table 4). On the final sampling date station 2 was significantly greater than station 4 but not stations 1 or 3. Stations 3 and 4 were not significantly different from the control or each other (Figs. 3 and 4; Table 4).

5.2.1.3 Genera of Chironomidae

Although significant differences were detectable in the family as a whole, apportioning them into their respective genera or species frequently yielded insufficient data for meaningful interpretation. A listing of those genera present in sufficient densities to compare among stations and points in time follow.

5.2.1.3.1 Paratendipes

Densities of this genus at station 2 were significantly greater than the control on all dates except 5 January, 1990 (Table 5). Through time the number of Paratendipes at station 2 declined steadily resulting in significantly lower densities on 24 November, 1989 and 5 January, 1990 , relative to 27 October, 1989 (Table 5).

5.2.1.3.2 Polypedilum illinoense

On the first three dates P. illinoese was only present at station 3 (Table 5). Interestingly, the temporal pattern at station 3 was similar to the pattern for total organisms in Chironomidae at station 2.

5.2.1.3.3 Polypedilum scalaenum and fallax

Both of these species were found primarily at station 2 (Table 5) and demonstrated the temporal trend characteristic of station 2 (e.g. A decrease on 10 November, 1989 and 24 November, 1989 and then a slight increase on 5 January, 1990). There was variability among these species regarding the significance of differences as compared to the control (Table 5).

5.2.1.3.4 Heleniella

Heleniella was found on all dates and was virtually exclusive to station 1 (Table 5).

5.2.1.3.5 Hydrobaenous

Hydrobaenous was recovered only at stations 1 and 3 during chlorination (Table 5). During the post-chlorination period they were also present at station 4. In fact they were most abundant at station 4 during recovery, although this did not become significantly different from the control until the final sampling date (Table 5).

5.2.1.3.6 Micropsectra

There was a significant increase in Micropsectra abundance at station 3 during recovery. This was the only station with appreciable numbers of Micropsectra (Table 5).

5.2.1.4 Ephemeroptera-A

Ephemeroptera-A refers to the early instar mayflies which were too small for further identification, and similar enough to be grouped together. The densities of Ephemeroptera-A (Table 4) differed from the other taxa. All stations had some Ephemeroptera-A during chlorination with the mean of station 2 being the largest (Note: the presence of only one specimen at station 3 precludes the use of statistical inference and, therefore, all subsequent statistical comparisons do not include station 3). Although the abundance of Ephemeroptera-A was too low for temporal differences to be significant the trend at station 2 was consistent with the

total organism plot. The changes during recovery at stations 1 and 4 differed only in magnitude (Table 4). A slight increase on 10 November, 1989 at the control leveled off for the remaining dates. At station 4, Ephemeroptera-A increased steadily for the first two post-chlorination dates and then returned to control levels on the final date. This increase was significantly different from the control on 24 November, 1989 (Table 4).

5.2.1.5 Hyalella azteca

The most prevelant crustacean was Hyalella azteca. This taxa, and the crustaceans in general, were only found at station 3. This prevents intersite tests of significance. Hyalella azteca did increase significantly on 5 January, 1990 as compared to 27 October, 1989 (Table 5).

5.2.1.6 Tubificidae

Tubificidae, the primary family in the order Oligocheta, did not vary significantly during the study, even at station 2. Stations 2 and 3 never differed from each other but were always significantly different from the control and station 4 (Table 5).

5.2.2 Relative Abundance

5.2.2.1 Total Organisms

In general the pattern of relative abundance was similar to absolute abundance (Table 6). The control and station 2 were always dominated by Chironomidae, with their contribution constituting between 50 and 70% of the total abundance. Stations 3 and 4,

however, varied more through time. Chironomidae and Oligochaeta were always prominent, but the tremendous increase in Crustacea on the final date relegated them to a lower standing based on percent abundance. Though there was a slight increase in the number of Chironomidae during non-chlorination, the relative abundance dropped from 44% during chlorination to 21% and 10% on the first and second post-chlorination dates, respectively (Table 6). This is the result of the increase in Ephemeroptera-A on 10 November, 1989 and 24 November, 1989. The return of Ephemeroptera-A to control levels coupled with the increase in Chironomidae lead to percent abundances on 5 January, 1990 of 21 and 71, respectively (Table 6).

5.2.2.2 Chironomidae

The family Chironomidae was dominated by Orthocladiinae on all dates at both the control and station 4 (Table 7). Orthocladiinae were also the major component of station 3, although Tanytarsini were somewhat more dominant on the second and final sampling dates. Chironominae were the dominant sub-family or tribe at station 2 for the first three dates (91-95%) and still the major sub-family (51%) on 5 January, 1990 with Orthocladiinae second at 37%.

The changes within the family Chironomidae during chlorination and on the final date during the post-chlorination period reveals some general trends. The control populations were never dominated by any one genus, however, Heleniella and Polypedilum fallax were the most abundant on both dates, with Orthocladius being equally dominant on the final date.

Station 2 changed from being predominantly Polypedilum

(scalaenum and fallax) during chlorination to include a second genus, Diplocladius, on the final non-chlorinated sampling date. Station 3 went from 33% Orthocladius to 33% Microspectra. Community composition at station 4, which was dominated by Orthocladius during chlorination, was more evenly distributed on the final sampling date with Diplocladius and Hydrobaenous being an equally large percentage (20%) of the community.

5.2.3 Taxa Richness (S)

5.2.3.1 Total Taxa Richness

Taxa refers to the lowest level of identification. Although the lowest level of identification varied by taxa it did not vary by date or station, thus allowing station by date comparisons. The total number of different taxa found at each station are in Table 8. During chlorination the control station is clearly the richest while station 4 is clearly the sparsest. Early in the post-chlorination period the macroinvertebrate communities at every station decreased in richness. On the final date the community at the control station was almost back to the original number of taxa. Taxa richness at the other 3 stations had increased in comparison to the taxa richness at each station during the chlorination period. The magnitude of this change increased with distance from the outfall with station 4 doubling in taxa richness. Total taxa richness for the family Chironomidae followed the same general trend but with a nearly 400% increase at station 4, and a more equitable richness for all stations on the final date (Table 9).

5.2.3.2 Average Taxa Richness

A 2-Way ANOVA of the average taxa richness per sample showed significant differences for date and station groupings but not for interactions. Again, as with total abundance, the appropriate tests were used for date and station comparisons. In general the average number of taxa per sample supports the total taxa richness data (Table 10). Specifically, station 4 was significantly lower than the control, as well as stations 2 and 3, during chlorination but not on the final recovery date. Station 4 did increase significantly on 5 January, 1990 with respect to the chlorination period (Table 10). This was also the case for Chironomidae richness. Taxa richness at station 1 did differ significantly across time (Fig. 5; Table 10). The small net increase through time at stations 2 and 3 were also not significant (Table 10).

5.2.4 Diversity

Shannon-Weiner Diversity was calculated for all organisms (Table 8) as well as the family Chironomidae (Table 9). The relative importance of Chironomidae at all stations on most sampling dates caused the two diversity indices to parallel each other.

5.2.4.1 Total Organisms

Station 1 was the most diverse station on all dates and station 2 the third most diverse on all dates. Stations three and four were the second most and the least diverse, respectively, on the first three sampling dates, but reversed places on the final date (Table 8). Stations 1 and 3 showed a small net decrease in Total Organisms

across time while stations 2 and 4 demonstrated a net increase in diversity with station 4 showing the greatest change. Interestingly the diversity at station 4 dropped on the first two post-chlorination dates.

5.2.4.2 Chironomidae

There were some notable differences in Chironomidae diversity as compared to total diversity. On the third sampling date station 3 was the most diverse and station 1 was the second most diverse (Fig. 6; Table 9). On the final sampling date the control had the highest diversity and the other three stations showed an increase in diversity with an increase in distance from the outfall. Also, station 4's diversity was the lowest on the first date, not the third, and highest on the final date. In fact, during chlorination, Chironomidae diversity at station 4 was the lowest of any station on any date (Fig. 7; Table 9).

5.2.5 Percent Community Similarity

The community structure at each station was compared to every other station on each date, and to the same station across time. The Percent Community Similarities derived from these comparisons are shown in tables 11 through 14.

5.2.5.1 Total Organisms

During chlorination community similarity to the control station decreased with distance from the outfall, from a high of 76% at station 2 to a low of 18% at station 4 (Table 11). During the

transition period (10 November, 1989 and 24 November, 1989) stations 2 and 3 fluctuated, with station 2 showing the greatest changes. During post-chlorination station 4 increased, decreased and then increased again to reach the highest level of similarity (82%) to the control of any station on any date. On the final date the similarity of stations 2 and 3 to the control were comparable to those during chlorination (Table 11).

Across time, the control remained 75-85% similar to the chlorination period (Table 12). The variability was greater at station 2 but the community on the last sampling date was 92% similar to the community on the first sampling date. Station 3 actually showed a decrease in similarity compared to the first date, from a high of 80% on the first two dates of recovery to a low of 59% on the final date. On 10 November, 1989 and 24 November, 1989 station 4 was clearly dissimilar to the first date it was, however, 80% similar to the chlorination period on the final date of recovery sampling (Table 12).

5.2.5.2 Chironomidae

Similar to Total Organisms, community similarity to the control station decreased with distance from the outfall, during chlorination, from a high of 30% at station 2 to a low of 18% at station 4 (Table 13). During the transition period (10 November, 1989 and 24 November, 1989) stations 2 and 3 fluctuated, with station 2 showing the greatest changes. During post-chlorination, station 4 increased, decreased and then increased again to reach the highest level of similarity (30%) to the control for that station. As

with Total Organisms the similarity of stations 2 and 3 to the control, on the final sampling date, were comparable to those during chlorination (Table 13).

Across time, the control remained 61-69% similar to the chlorination period (Table 14). At station 2 similarity decreased across time and the community on the last sampling date was only 59% similar to the community on the first sampling date. Station 3 also showed a decrease in similarity compared to the first date, especially on the final date. Station 4 was clearly the most dissimilar, as compared to the first date, on all subsequent sampling dates (Table 14).

Table 1. Sediment percent composition.

Sampling Date / Station	Diameter (mm) > 6.3	2.0 - 6.3	0.500 - 2.0	0.075 - 0.500	< 0.075
10/27/89					
Station 1	22.0 %	22.3 %	26.6 %	28.3 %	2.7 %
Station 2	40.3 %	23.9 %	18.8 %	16.6 %	0.4 %
Station 3	35.7 %	27.3 %	20.0 %	15.1 %	1.9 %
Station 4	11.4 %	51.1 %	28.5 %	8.9 %	0.0 %
01/05/90					
Station 1	17.5 %	16.7 %	31.2 %	32.8 %	2.1 %
Station 2	27.5 %	32.4 %	20.2 %	19.1 %	0.7 %
Station 3	34.2 %	20.1 %	19.5 %	23.7 %	2.5 %
Station 4	5.4 %	47.4 %	33.7 %	13.2 %	0.4 %

Table 2. Sediment Percent Similarity of stations two, three, and four relative to the control (station 1) for that sampling date.

	Sampling Date	
Station	10/27/89	01/05/90
Two	80 %	74 %
Three	81 %	79 %
Four	69 %	67 %

Table 3. Sediment Percent Similarity of stations one and four during recovery relative to themselves on 27 October, 1989 (Chlorination).

	Station	
Sampling Date	1	4
1/5/90	89 %	90 %

Table 4-a. Mean number (± 1.96*SE) per core of the major taxonomic groups. Based on 20 core samples collected on 27 October, 1989.

	Station			
Taxa	1	2	3	4
Total Organisms	11.10 ± 2.64	48.30 ± 15.75	20.30 ± 4.79	2.40 ± 0.73
Coleoptera	1.80 ± 0.63	0.20 ± 0.18	0.00 ± 0.00	0.00 ± 0.00
Crustacea	0.00 ± 0.00	0.00 ± 0.00	2.45 ± 1.10	0.00 ± 0.00
Diptera*	0.40 ± 0.26	1.65 ± 0.67	0.50 ± 0.27	0.15 ± 0.16
Chironomidae	6.55 ± 1.96	31.40 ± 11.56	6.65 ± 2.37	1.05 ± 0.48
Ephemeroptera	0.65 ± 0.38	3.85 ± 3.28	0.10 ± 0.20	0.45 ± 0.44
Ephemeroptera-A	0.65 ± 0.38	3.85 ± 3.28	0.05 ± 0.10	0.45 ± 0.44
Hirudinae	0.00 ± 0.00	0.00 ± 0.00	0.45 ± 0.30	0.00 ± 0.00
Mollusca	0.00 ± 0.00	0.00 ± 0.00	0.25 ± 0.19	0.05 ± 0.10
Nematatoda	0.30 ± 0.25	5.90 ± 5.76	3.20 ± 1.57	0.00 ± 0.00
Odonata	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Oligochetae	0.70 ± 0.55	5.25 ± 1.75	6.70 ± 2.63	0.50 ± 0.46
Plecoptera	0.50 ± 0.27	0.00 ± 0.00	0.00 ± 0.00	0.20 ± 0.18
Trichoptera	0.15 ± 0.16	0.05 ± 0.10	0.00 ± 0.00	0.00 ± 0.00
Unidentified	0.05 ± 0.10	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00

* Not including Chironomidae.
Ephemeroptera-A = Early instar mayflies.

Table 4-b. Mean number (± 1.96*SE) per core of the major taxonomic groups. Based on 11 core samples collected on 10 November, 1989.

	Station			
Taxa	1	2	3	4
Total Organisms	13.00 ± 5.16	20.45 ± 10.50	21.09 ± 8.28	12.27 ± 5.86
Coleoptera	1.67 ± 0.77	0.33 ± 0.24	0.17 ± 0.18	0.00 ± 0.00
Crustacea	0.00 ± 0.00	0.00 ± 0.00	4.73 ± 2.68	0.27 ± 0.38
Diptera*	0.82 ± 0.69	1.64 ± 1.80	0.18 ± 0.36	0.00 ± 0.00
Chironomidae	6.55 ± 2.54	13.82 ± 7.87	8.00 ± 3.54	2.55 ± 2.51
Ephemeroptera	6.67 ± 2.57	0.83 ± 0.48	0.00 ± 0.00	16.33 ± 4.60
Ephemeroptera-A	3.64 ± 2.57	0.45 ± 0.48	0.00 ± 0.00	8.91 ± 4.60
Hirudinae	0.00 ± 0.00	0.00 ± 0.00	0.18 ± 0.36	0.00 ± 0.00
Mollusca	0.00 ± 0.00	0.09 ± 0.18	0.91 ± 0.85	0.09 ± 0.18
Nematatoda	0.36 ± 0.55	0.09 ± 0.18	1.73 ± 1.18	0.00 ± 0.00
Odonata	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Oligochetae	0.18 ± 0.36	4.18 ± 2.36	5.27 ± 2.91	0.00 ± 0.00
Plecoptera	0.27 ± 0.28	0.00 ± 0.00	0.00 ± 0.00	0.36 ± 0.30
Trichoptera	0.27 ± 0.28	0.00 ± 0.00	0.00 ± 0.00	0.09 ± 0.18
Unidentified	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00

* Not including Chironomidae.
Ephemeroptera-A = Early instar mayflies.

Table 4-c. Mean number (± 1.96*SE) per core of the major taxonomic groups. Based on 11 core samples collected on 24 November, 1989.

Taxa	Station 1	Station 2	Station 3	Station 4
Total Organisms	15.36 ± 6.60	18.55 ± 4.84	20.09 ± 4.79	20.00 ± 9.61
Coleoptera	3.33 ± 1.32	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Crustacea	0.09 ± 0.18	0.00 ± 0.00	4.55 ± 1.72	0.00 ± 0.00
Diptera*	0.45 ± 0.41	0.27 ± 0.38	0.00 ± 0.00	0.00 ± 0.00
Chironomidae	9.00 ± 4.27	11.00 ± 4.33	10.00 ± 2.47	2.09 ± 1.07
Ephemeroptera	4.17 ± 1.67	1.67 ± 0.67	0.00 ± 0.00	32.33 ± 9.24
Ephemeroptera-A	2.27 ± 1.67	0.91 ± 0.67	0.00 ± 0.00	17.64 ± 9.24
Hirudinae	0.00 ± 0.00	0.00 ± 0.00	0.18 ± 0.24	0.00 ± 0.00
Mollusca	0.00 ± 0.00	0.00 ± 0.00	0.27 ± 0.53	0.00 ± 0.00
Nematatoda	0.45 ± 0.41	0.55 ± 0.55	0.18 ± 0.24	0.00 ± 0.00
Odonata	0.00 ± 0.00	0.09 ± 0.18	0.00 ± 0.00	0.00 ± 0.00
Oligochetae	0.36 ± 0.40	5.73 ± 3.44	4.91 ± 2.17	0.00 ± 0.00
Plecoptera	0.91 ± 0.62	0.00 ± 0.00	0.00 ± 0.00	0.27 ± 0.28
Trichoptera	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Unidentified	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00

* Not including Chironomidae.
Ephemeroptera-A = Early instar mayflies.

Table 4-d. Mean number (± 1.96*SE) per core of the major taxonomic groups. Based on 11 core samples collected on 5 January, 1990.

	Station			
Taxa	1	2	3	4
Total Organisms	13.64 ± 6.67	35.55 ± 10.49	51.64 ± 22.24	14.18 ± 6.27
Coleoptera	2.33 ± 0.80	0.33 ± 0.24	0.00 ± 0.00	0.00 ± 0.00
Crustacea	0.09 ± 0.18	0.00 ± 0.00	27.73 ± 15.69	0.18 ± 0.24
Diptera*	0.36 ± 0.40	2.45 ± 1.59	0.64 ± 0.55	0.27 ± 0.28
Chironomidae	9.64 ± 5.46	24.18 ± 8.80	14.18 ± 7.04	10.09 ± 4.78
Ephemeroptera	2.17 ± 1.58	3.50 ± 1.31	0.00 ± 0.00	5.50 ± 2.16
Ephemeroptera-A	1.09 ± 1.60	1.91 ± 1.31	0.00 ± 0.00	2.91 ± 2.15
Hirudinae	0.00 ± 0.00	0.00 ± 0.00	0.64 ± 0.40	0.00 ± 0.00
Mollusca	0.00 ± 0.00	0.00 ± 0.00	0.55 ± 0.55	0.00 ± 0.00
Nematatoda	0.27 ± 0.28	2.36 ± 3.28	2.55 ± 2.59	0.18 ± 0.24
Odonata	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Oligochetae	0.09 ± 0.18	4.18 ± 1.29	5.36 ± 2.43	0.27 ± 0.28
Plecoptera	0.55 ± 0.31	0.09 ± 0.18	0.00 ± 0.00	0.09 ± 0.18
Trichoptera	0.09 ± 0.18	0.18 ± 0.24	0.00 ± 0.00	0.00 ± 0.00
Unidentified	0.09 ± 0.30	0.00 ± 0.00	0.00 ± 0.00	0.09 ± 0.30

* Not including Chironomidae.
Ephemeroptera-A = Early instar mayflies.

Table 5-a. Mean number (± 1.96*SE) per core of selected taxa. Based on 20 core samples collected on 27 October, 1989.

Taxa	Station 1	Station 2	Station 3	Station 4
Crustacea				
Hyalella azteca	0.00 ± 0.00	0.00 ± 0.00	0.95 ± 0.44	0.00 ± 0.00
Diptera				
Heleniella sp.	1.25 ± 0.68	0.05 ± 0.10	0.00 ± 0.00	0.00 ± 0.00
Hydrobaenus sp.	0.10 ± 0.13	0.00 ± 0.00	0.35 ± 0.26	0.00 ± 0.00
Micropsectra sp.	0.20 ± 0.18	0.00 ± 0.00	0.20 ± 0.23	0.00 ± 0.00
Paratendipes sp.	0.15 ± 0.21	2.66 ± 1.14	0.10 ± 0.13	0.00 ± 0.00
Polypedilum illinoense	0.00 ± 0.00	0.00 ± 0.00	1.40 ± 0.69	0.00 ± 0.00
Polypedilum scalaenum	0.55 ± 0.44	16.36 ± 6.51	0.05 ± 0.10	0.00 ± 0.00
Polypedilum fallax	0.95 ± 0.50	9.56 ± 4.18	0.15 ± 0.16	0.00 ± 0.00
Oligochaeta				
Tubififcidae	0.50 ± 0.52	4.80 ± 1.77	6.30 ± 2.69	0.10 ± 0.20

Table 5-b. Mean number (± 1.96*SE) per core of selected taxa. Based on 11 core samples collected on 10 November, 1989.

	Station			
Taxa	1	2	3	4
Crustacea				
Hyalella azteca	0.00 ± 0.00	0.00 ± 0.00	2.09 ± 1.33	0.00 ± 0.00
Diptera				
Heleniella sp.	0.91 ± 0.67	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Hydrobaenus sp.	0.18 ± 0.24	0.00 ± 0.00	0.47 ± 0.58	1.73 ± 1.93
Micropsectra sp.	0.09 ± 0.18	0.00 ± 0.00	3.02 ± 1.55	0.00 ± 0.00
Paratendipes sp.	0.27 ± 0.28	1.60 ± 0.90	0.09 ± 0.18	0.00 ± 0.00
Polypedilum illinoense	0.00 ± 0.00	0.00 ± 0.00	0.45 ± 0.31	0.00 ± 0.00
Polypedilum scalaenum	0.82 ± 0.52	6.67 ± 3.30	0.00 ± 0.00	0.09 ± 0.18
Polypedilum fallax	1.09 ± 0.89	4.16 ± 4.49	0.10 ± 0.19	0.09 ± 0.18
Oligochaeta				
Tubififcidae	0.33 ± 0.36	7.50 ± 2.36	8.17 ± 2.50	0.00 ± 0.00

Table 5-c. Mean number (± 1.96*SE) per core of selected taxa. Based on 11 core samples collected on 24 November, 1989.

Taxa	Station 1	Station 2	Station 3	Station 4
Crustacea				
Hyalella azteca	0.00 ± 0.00	0.00 ± 0.00	1.64 ± 1.30	0.00 ± 0.00
Diptera				
Heleniella sp.	1.45 ± 0.93	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Hydrobaenus sp.	0.00 ± 0.00	0.09 ± 0.18	0.55 ± 0.48	1.55 ± 0.89
Micropsectra sp.	0.00 ± 0.00	0.09 ± 0.18	2.82 ± 1.32	0.00 ± 0.00
Paratendipes sp.	0.00 ± 0.00	0.85 ± 0.45	0.27 ± 0.28	0.00 ± 0.00
Polypedilum Illinoense	0.00 ± 0.00	0.00 ± 0.00	0.18 ± 0.24	0.00 ± 0.00
Polypedilum scalaenum	0.64 ± 0.55	4.99 ± 1.63	0.09 ± 0.18	0.00 ± 0.00
Polypedilum fallax	1.68 ± 0.62	4.20 ± 2.46	0.55 ± 0.48	0.00 ± 0.00
Oligochaeta				
Tubififcidae	0.50 ± 0.38	8.33 ± 2.66	7.83 ± 1.91	0.00 ± 0.00

Table 5-d. Mean number (± 1.96*SE) per core of selected taxa. Based on 11 core samples collected on 5 January, 1990.

	Station							
Taxa	1		2		3		4	
Crustacea								
Hyalella azteca	0.00	± 0.00	0.00	± 0.00	5.73	± 3.86	0.00	± 0.00
Diptera								
Heleniella sp.	2.07	± 1.81	0.00	± 0.00	0.00	± 0.00	0.00	± 0.00
Hydrobaenus sp.	0.30	± 0.31	0.00	± 0.00	0.09	± 0.18	2.38	± 1.58
Micropsectra sp.	0.11	± 0.22	0.09	± 0.18	4.72	± 3.08	0.60	± 0.49
Paratendipes sp.	0.21	± 0.28	0.46	± 0.51	0.31	± 0.32	0.57	± 0.33
Polypedilum Illinoense	0.21	± 0.28	0.26	± 0.52	0.89	± 0.83	0.09	± 0.18
Polypedilum scalaenum	0.79	± 0.95	6.55	± 4.75	0.27	± 0.38	0.00	± 0.00
Polypedilum fallax	1.32	± 0.86	6.14	± 3.02	0.09	± 0.18	0.09	± 0.18
Oligochaeta								
Tubififcidae	0.17	± 0.18	7.33	± 1.27	7.33	± 2.05	0.00	± 0.00

Table 6-a. Total and relative abundance of the major taxonomic groups. Collected at each station on 27 October, 1989. Based on 20 core samples.

	Station							
	1		2		3		4	
Taxa	Total	%	Total	%	Total	%	Total	%
Total Organisms	222		966		406		48	
Coleoptera	36	16.22	4	0.41	0	0.00	0	0.00
Crustacea	0	0.00	0	0.00	49	12.07	0	0.00
Diptera *	8	3.60	34	3.52	11	2.71	3	6.25
Chironomidae	131	59.01	627	64.91	132	32.51	21	43.75
EPT	26	11.71	78	8.07	2	0.49	13	27.08
Epemeroptera	13	5.86	77	7.97	2	0.49	9	18.75
Plecoptera	10	4.50	0	0.00	0	0.00	4	8.33
Trichoptera	3	1.35	1	0.10	0	0.00	0	0.00
Hirudinae	0	0.00	0	0.00	9	2.22	0	0.00
Mollusca	0	0.00	0	0.00	5	1.23	1	2.08
Nematoda	6	2.70	118	12.22	64	15.76	0	0.00
Odonata	0	0.00	0	0.00	0	0.00	0	0.00
Oligochaeta	14	6.31	105	10.87	134	33.00	10	20.83
Unidentifiable	1	0.45	0	0.00	0	0.00	0	0.00

* Not including Chironomidae.
EPT = The sum of the orders Epemeroptera, Plecoptera, and Trichoptera.

Table 6-b. Total and relative abundance of the major taxonomic groups. Collected at each station on 10 November, 1989. Based on 11 core samples.

	Station							
	1		2		3		4	
Taxa	Total	%	Total	%	Total	%	Total	%
Total Organisms	143		225		232		135	
Coleoptera	10	6.99	2	0.89	1	0.43	0	0.00
Crustacea	0	0.00	0	0.00	52	22.41	3	2.22
Diptera *	9	6.29	18	8.00	2	0.86	0	0.00
Chironomidae	72	50.35	152	67.56	88	37.93	28	20.74
EPT	46	32.17	5	2.22	0	0.00	103	76.30
Epemeroptera	40	27.97	5	2.22	0	0.00	98	72.59
Plecoptera	3	2.10	0	0.00	0	0.00	4	2.96
Trichoptera	3	2.10	0	0.00	0	0.00	1	0.74
Hirudinae	0	0.00	0	0.00	2	0.86	0	0.00
Mollusca	0	0.00	1	0.44	10	4.31	1	0.74
Nematoda	4	2.80	1	0.44	19	8.19	0	0.00
Odonata	0	0.00	0	0.00	0	0.00	0	0.00
Oligochaeta	2	1.40	46	20.44	58	25.00	0	0.00
Unidentifiable	0	0.00	0	0.00	0	0.00	0	0.00

* Not including Chironomidae.
EPT = The sum of the orders Epemeroptera, Plecoptera, and Trichoptera.

Table 6-c. Total and relative abundance of the major taxonomic groups. Collected at each station on 24 November, 1989. Based on 11 core samples.

	Station							
	1		2		3		4	
Taxa	Total	%	Total	%	Total	%	Total	%
Total Organisms	169		204		221		220	
Coleoptera	20	11.83	0	0.00	0	0.00	0	0.00
Crustacea	1	0.59	0	0.00	50	22.62	0	0.00
Diptera *	5	2.96	3	1.47	1	0.45	0	0.00
Chironomidae	99	58.58	121	59.31	109	49.32	23	10.45
EPT	35	20.71	10	4.90	0	0.00	197	89.55
Epemeroptera	25	14.79	10	4.90	0	0.00	194	88.18
Plecoptera	10	5.92	0	0.00	0	0.00	3	1.36
Trichoptera	0	0.00	0	0.00	0	0.00	0	0.00
Hirudinae	0	0.00	0	0.00	2	0.90	0	0.00
Mollusca	0	0.00	0	0.00	3	1.36	0	0.00
Nematoda	5	2.96	6	2.94	2	0.90	0	0.00
Odonata	0	0.00	1	0.49	0	0.00	0	0.00
Oligochaeta	4	2.37	63	30.88	54	24.43	0	0.00
Unidentifiable	0	0.00	0	0.00	0	0.00	0	0.00

* Not including Chironomidae.
EPT = The sum of the orders Epemeroptera, Plecoptera, and Trichoptera.

Table 6-d. Total and relative abundance of the major taxonomic groups. Collected at each station on 5 January, 1990. Based on 11 core samples.

	Station							
	1		2		3		4	
Taxa	Total	%	Total	%	Total	%	Total	%
Total Organisms	150		391		568		156	
Coleoptera	14	9.33	2	0.51	0	0.00	0	0.00
Crustacea	1	0.67	0	0.00	305	53.70	2	1.28
Diptera *	4	2.67	27	6.91	7	1.23	3	1.92
Chironomidae	106	70.67	266	68.03	156	27.46	111	71.15
EPT	20	13.33	24	6.14	0	0.00	34	21.79
Epemeroptera	13	8.67	21	5.37	0	0.00	33	21.15
Plecoptera	6	4.00	1	0.26	0	0.00	1	0.64
Trichoptera	1	0.67	2	0.51	0	0.00	0	0.00
Hirudinae	0	0.00	0	0.00	7	1.23	0	0.00
Mollusca	0	0.00	0	0.00	6	1.06	0	0.00
Nematoda	3	2.00	26	6.65	28	4.93	2	1.28
Odonata	0	0.00	0	0.00	0	0.00	0	0.00
Oligochaeta	1	0.67	46	11.76	59	10.39	3	1.92
Unidentifiable	1	0.67	0	0.00	0	0.00	1	0.64

* Not including Chironomidae.
EPT = The sum of the orders Epemeroptera, Plecoptera, and Trichoptera.

Table 7. Relative abundance of selected sub-families and tribes of the family Chironomidae .

Sample Date / Taxa	Station 1	2	3	4
10/27/89				
Chironominae	28.24 %	94.99 %	34.09 %	0.00 %
Orthocladiinae	32.82 %	1.35 %	49.24 %	80.95 %
Tanypodinae	20.61 %	2.94 %	3.03 %	4.76 %
Tanytarsini	9.92 %	0.55 %	12.12 %	9.52 %
Other	8.40 %	0.17 %	1.52 %	4.76 %
11/10/89				
Chironominae	36.11 %	91.97 %	16.08 %	7.14 %
Orthocladiinae	45.83 %	3.29 %	33.64 %	85.71 %
Tanypodinae	13.89 %	3.95 %	3.41 %	0.00 %
Tanytarsini	4.17 %	0.79 %	46.88 %	7.14 %
Other	0.00 %	0.00 %	0.00 %	0.00 %
11/24/89				
Chironominae	27.78 %	93.10 %	12.84 %	4.35 %
Orthocladiinae	48.48 %	3.43 %	40.37 %	91.30 %
Tanypodinae	17.68 %	0.00 %	10.09 %	0.00 %
Tanytarsini	3.03 %	1.82 %	36.70 %	4.35 %
Other	3.03 %	1.65 %	0.00 %	0.00 %
1/5/90				
Chironominae	32.59 %	57.58 %	12.96 %	9.28 %
Orthocladiinae	53.02 %	37.44 %	33.84 %	64.23 %
Tanypodinae	12.03 %	2.63 %	7.83 %	3.87 %
Tanytarsini	1.18 %	0.92 %	45.38 %	19.91 %
Other	1.18 %	1.43 %	0.00 %	2.70 %

Table 8. Diversity and total taxa richness. Total taxa richness is the number of different taxa per site.

Sample Date / Parameter	N	Station 1	2	3	4
10/27/89					
Diversity H'	20	3.23	2.06	2.47	2.01
Taxa Richness S		42	29	33	14
11/10/89					
Diversity H'	11	2.66	1.97	2.52	1.07
Taxa Richness S		29	20	25	12
11/24/89					
Diversity H'	11	2.77	1.99	2.53	0.52
Taxa Richness S		25	19	27	10
1/5/90					
Diversity H'	11	3.10	2.35	2.27	2.65
Taxa Richness S		37	32	38	29

Table 9. Diversity and total taxa richness for the family Chironomidae. Total taxa richness is the number of different taxa per site.

		Station			
Sample Date / Parameter	N	1	2	3	4
10/27/89					
Diversity H'	20	2.72	1.33	2.31	0.75
Taxa Richness S		25	17	20	5
11/10/89					
Diversity H'	11	2.31	1.37	1.81	1.09
Taxa Richness S		17	10	14	6
11/24/89					
Diversity H'	11	2.24	1.34	2.27	1.04
Taxa Richness S		14	11	20	7
1/5/90					
Diversity H'	11	2.55	1.75	2.32	2.40
Taxa Richness S		21	21	22	19

Table 10. Average taxa richness (± 1.96 * SE) of Total Organisms and Chironomidae per core. Based on 20 core samples for 10/27/89 and 11 core samples for the remaining dates.

	Station			
Sample Date / Taxa	1	2	3	4
10/27/89				
Total Organisms	7.35 ± 1.38	7.55 ± 0.97	8.30 ± 1.74	2.50 ± 0.89
Chironomidae	4.15 ± 1.09	3.95 ± 0.52	4.05 ± 1.15	0.75 ± 0.31
11/10/89				
Total Organisms	7.00 ± 1.83	5.46 ± 1.66	8.09 ± 1.99	2.91 ± 1.20
Chironomidae	4.46 ± 1.23	3.46 ± 1.14	4.00 ± 1.19	1.18 ± 0.88
11/24/89				
Total Organisms	8.00 ± 1.92	5.55 ± 1.20	8.18 ± 1.65	2.55 ± 0.67
Chironomidae	4.64 ± 1.48	3.46 ± 0.61	5.18 ± 1.31	1.27 ± 0.54
1/5/90				
Total Organisms	7.73 ± 3.14	9.00 ± 1.23	11.55 ± 2.65	7.64 ± 1.72
Chironomidae	4.91 ± 2.00	5.36 ± 0.96	5.91 ± 2.02	5.73 ± 1.77

Table 11. Total Organisms Percent Community Similarity of stations two, three, and four relative to the control (station 1) for that sampling date.

	Sampling Date			
Station	10/27/89	11/10/89	11/24/89	01/05/90
Two	76 %	65 %	39 %	79 %
Three	54 %	50 %	57 %	53 %
Four	18 %	64 %	49 %	82 %

Table 12. Total Organisms Percent Community Similarity of each station during the non-chlorination period relative to its self on 10/27/89 (chorination period).

	Station			
Sampling Date	1	2	3	4
11/10/89	78 %	83 %	80 %	53 %
11/24/89	87 %	53 %	80 %	46 %
01/05/90	85 %	92 %	59 %	79 %

Table 13. Chironomidae Percent Community Similarity of stations two, three, and four relative to the control (station 1) for that sampling date.

	Sampling Date			
Station	10/27/89	11/10/89	11/24/89	01/05/90
Two	30 %	40 %	32 %	33 %
Three	27 %	40 %	40 %	29 %
Four	18 %	26 %	11 %	30 %

Table 14. Chironomidae Percent Community Similarity of each station during the non-chlorination period relative to its self on 10/27/89 (chorination period).

	Station			
Sampling Date	1	2	3	4
11/10/89	61 %	92 %	56 %	19 %
11/24/89	65 %	86 %	52 %	4 %
01/05/90	69 %	59 %	39 %	15 %

Figure 1. Mean number (± 1.96*S.E.) of total organisms per core at each station on all four sampling dates. The first date, 27 October, 1989, is three days before the termination of chlorination. Sampling dates 2, 3, and 4 are 11, 25, and 67 days after the termination of chlorination, respectively.Station 1 (control) is located 80 meters above the outfall and stations 2, 3, and 4 are located 80, 860, and 3700 meters below the outfall, respectively.

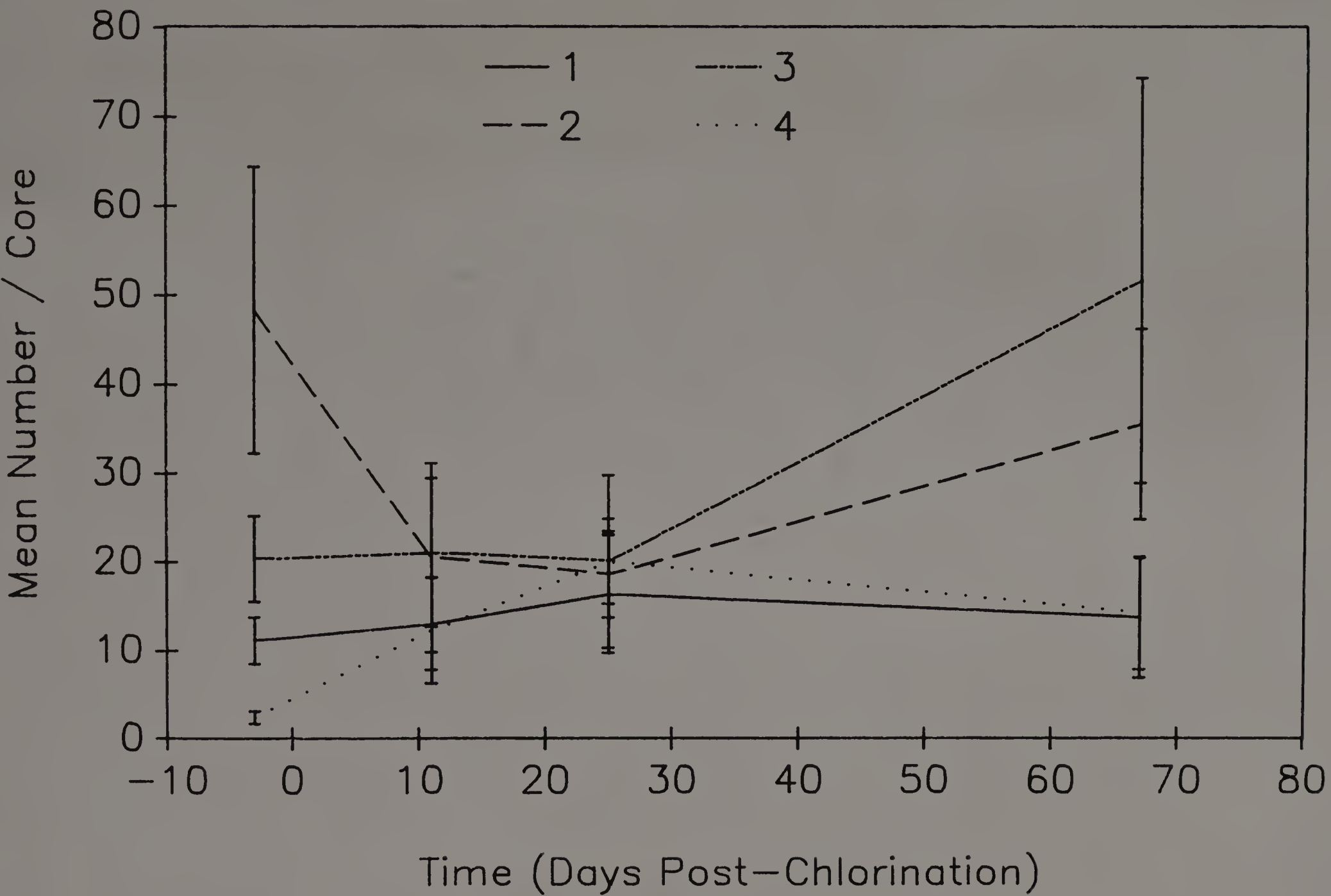

1
2
3
4
Mean Number / Core
0
10
20
30
40
50
60
70
80
−10
0
10
20
30
40
50
60
70
80
Time (Days Post−Chlorination)

Figure 2. Mean number (± 1.96*S.E.) of total organisms per core at each station during the chlorination and post-chlorination periods. "No Chlorine" data based on 20 samples collected on 5 January, 1990. "Chlorine" data based on 11 samples collected on 27 October, 1989. Station 1 (control) is located 80 meters above the outfall and stations 2, 3, and 4 are located 80, 860, and 3700 meters below the outfall, respectively.

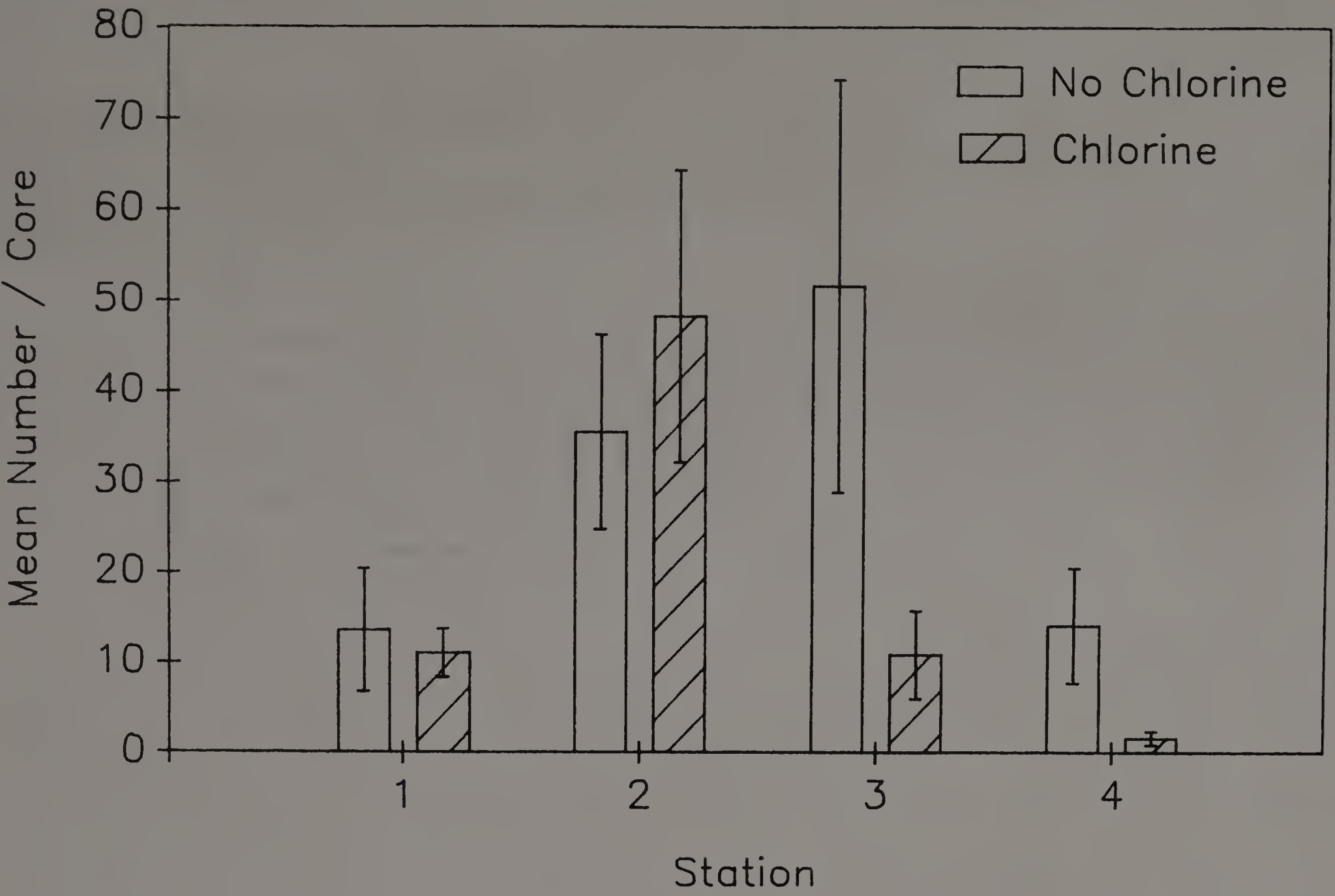

Mean Number / Core
80
70
60
50
40
30
20
10
0
1
2
3
4
Station
No Chlorine
Chlorine

Figure 3. Mean number (± 1.96*S.E.) of Chironomidae per core at each station on all four sampling dates. The first date, 27 October, 1989, is three days before the termination of chlorination. Sampling dates 2, 3, and 4 are 11, 25, and 67 days after the termination of chlorination, respectively.Station 1 (control) is located 80 meters above the outfall and stations 2, 3, and 4 are located 80, 860, and 3700 meters below the outfall, respectively.

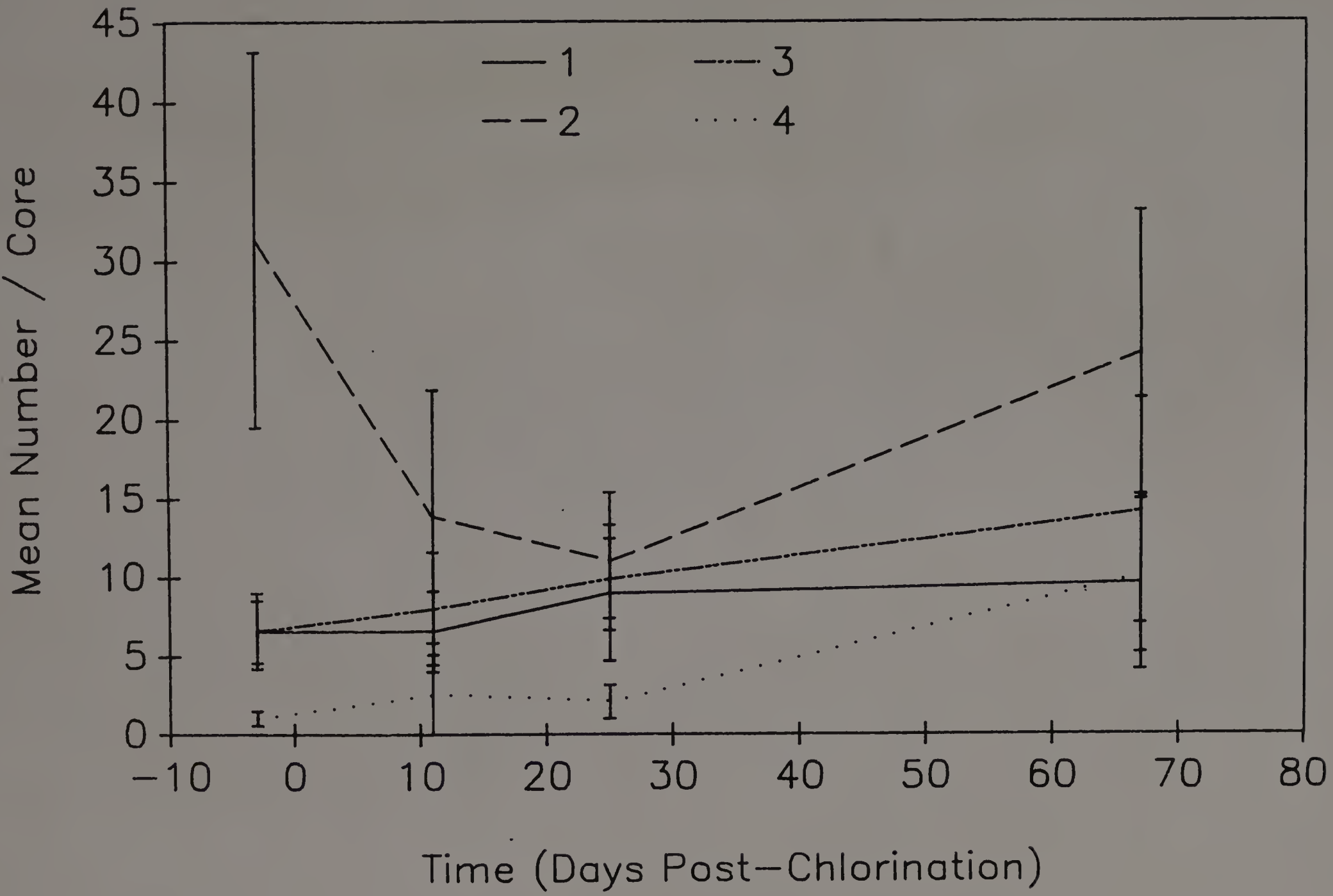

45
40
35
30
25
20
15
10
5
0
Mean Number / Core
1
2
3
4
−10
0
10
20
30
40
50
60
70
80
Time (Days Post−Chlorination)

Figure 4. Mean number (± 1.96*S.E.) of Chironomidae per core at each station during the chlorination and post-chlorination periods. "No Chlorine" data based on 20 samples collected on 5 January, 1990. "Chlorine" data based on 11 samples collected on 27 October, 1989. Station 1 (control) is located 80 meters above the outfall and stations 2, 3, and 4 are located 80, 860, and 3700 meters below the outfall, respectively.

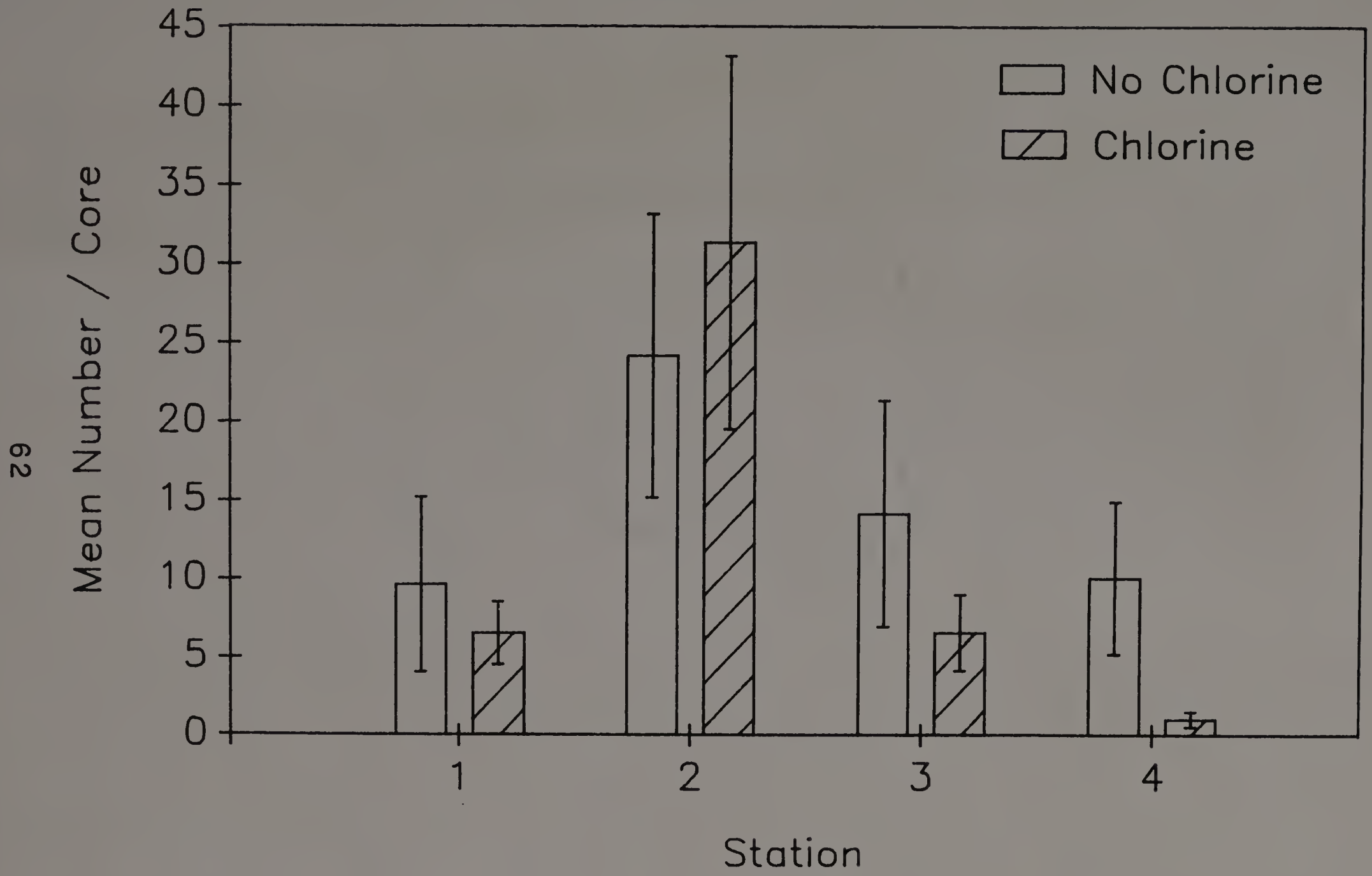

No Chlorine
Chlorine
Mean Number / Core
0
5
10
15
20
25
30
35
40
45
1
2
3
4
Station

Figure 5. Mean number (± 1.96*S.E.) of different Chironomidae taxa (genera and species) per core at each station during the chlorination and post-chlorination periods. "No Chlorine" data based on 20 samples collected on 5 January, 1990. "Chlorine" data based on 11 samples collected on 27 October, 1989. Station 1 (control) is located 80 meters above the outfall and stations 2, 3, and 4 are located 80, 860, and 3700 meters below the outfall, respectively.

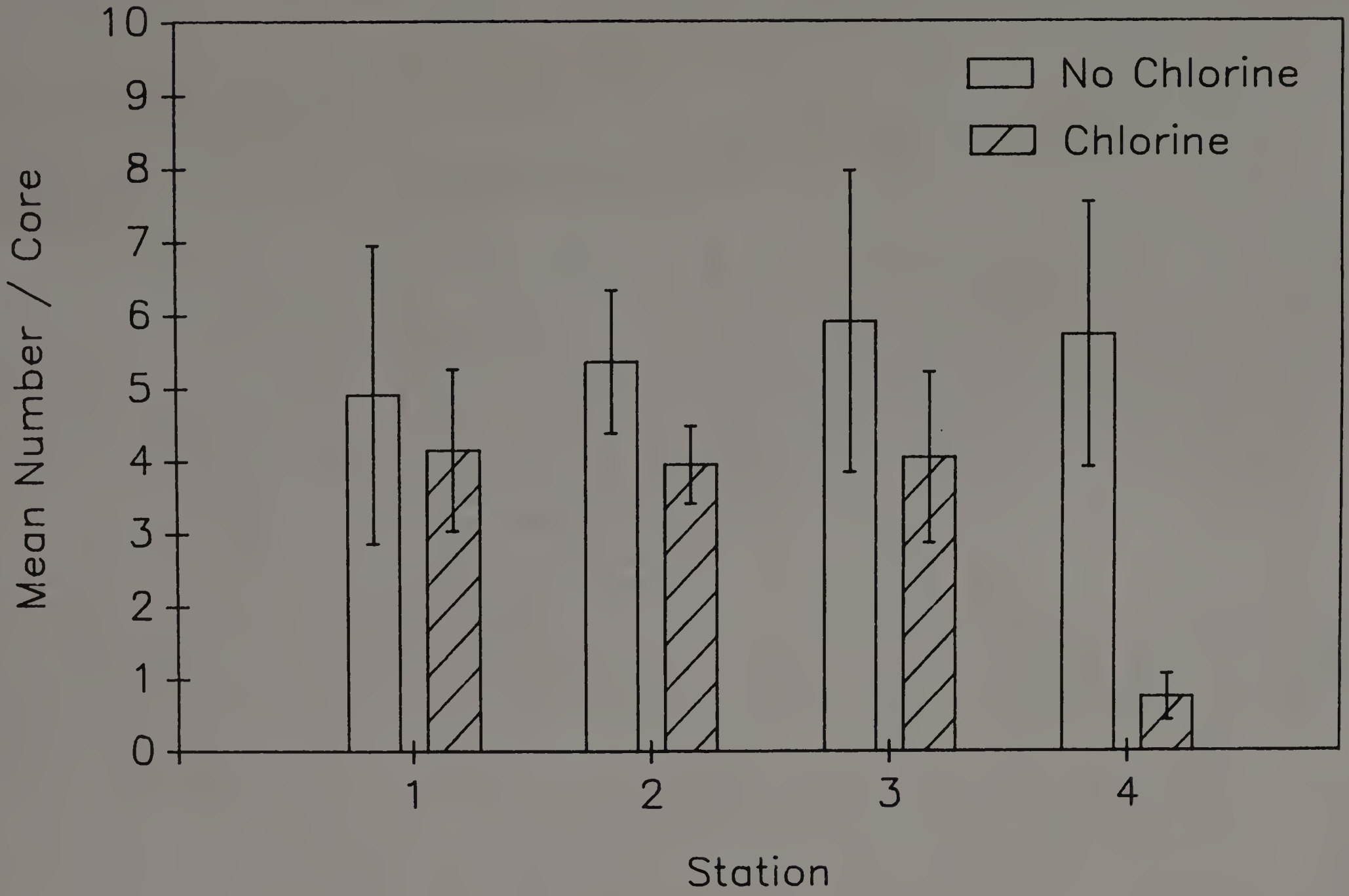

No Chlorine
Chlorine
Mean Number / Core
0
1
2
3
4
5
6
7
8
9
10
1
2
3
4
Station

Figure 6. Chironomidae diversity (Shannon-Wiener) at each station on all four sampling dates. The first date, 27 October, 1989, is three days before the termination of chlorination. Sampling dates 2, 3, and 4 are 11, 25, and 67 days after the termination of chlorination, respectively.Station 1 (control) is located 80 meters above the outfall and stations 2, 3, and 4 are located 80, 860, and 3700 meters below the outfall, respectively.

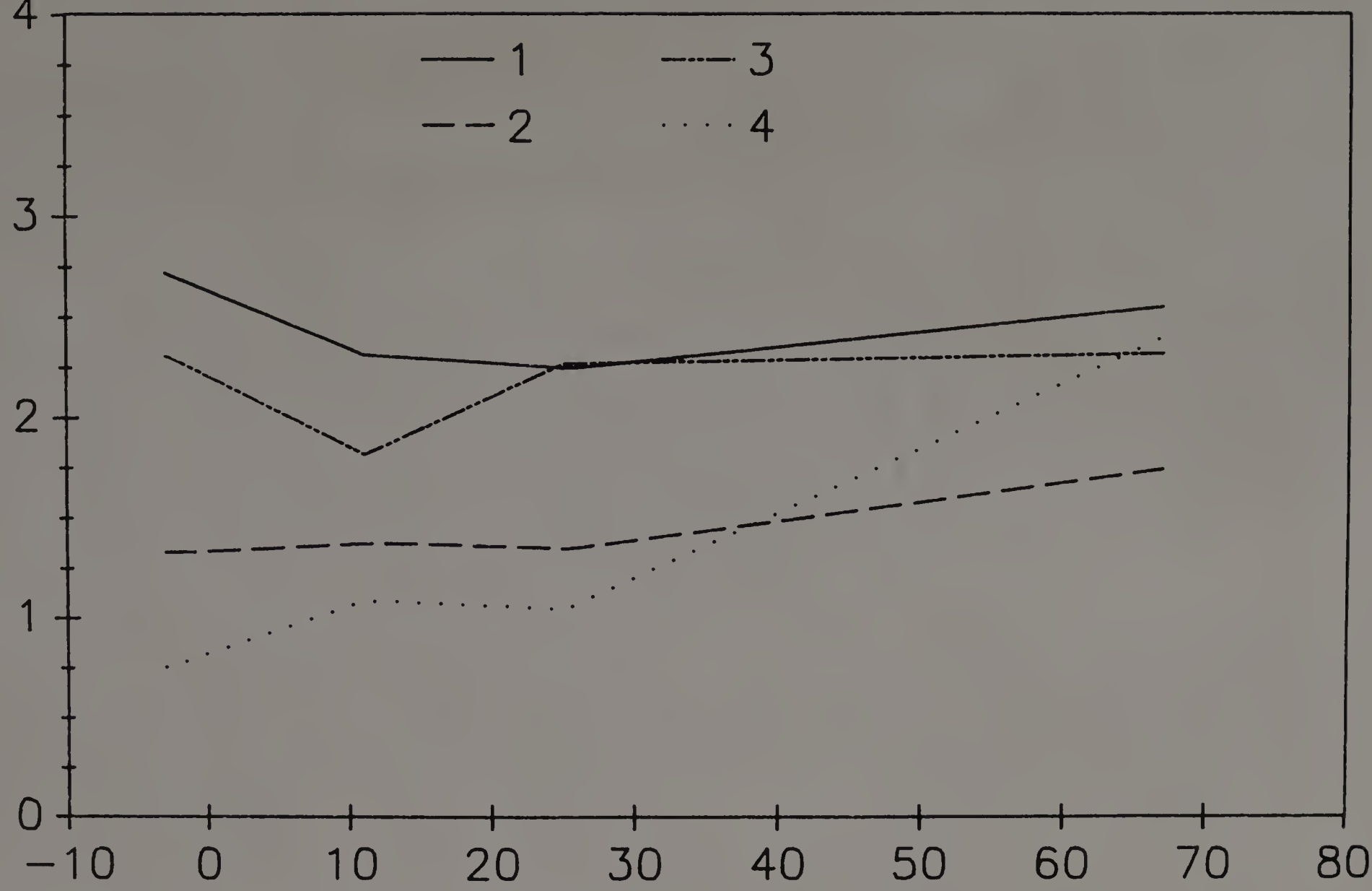
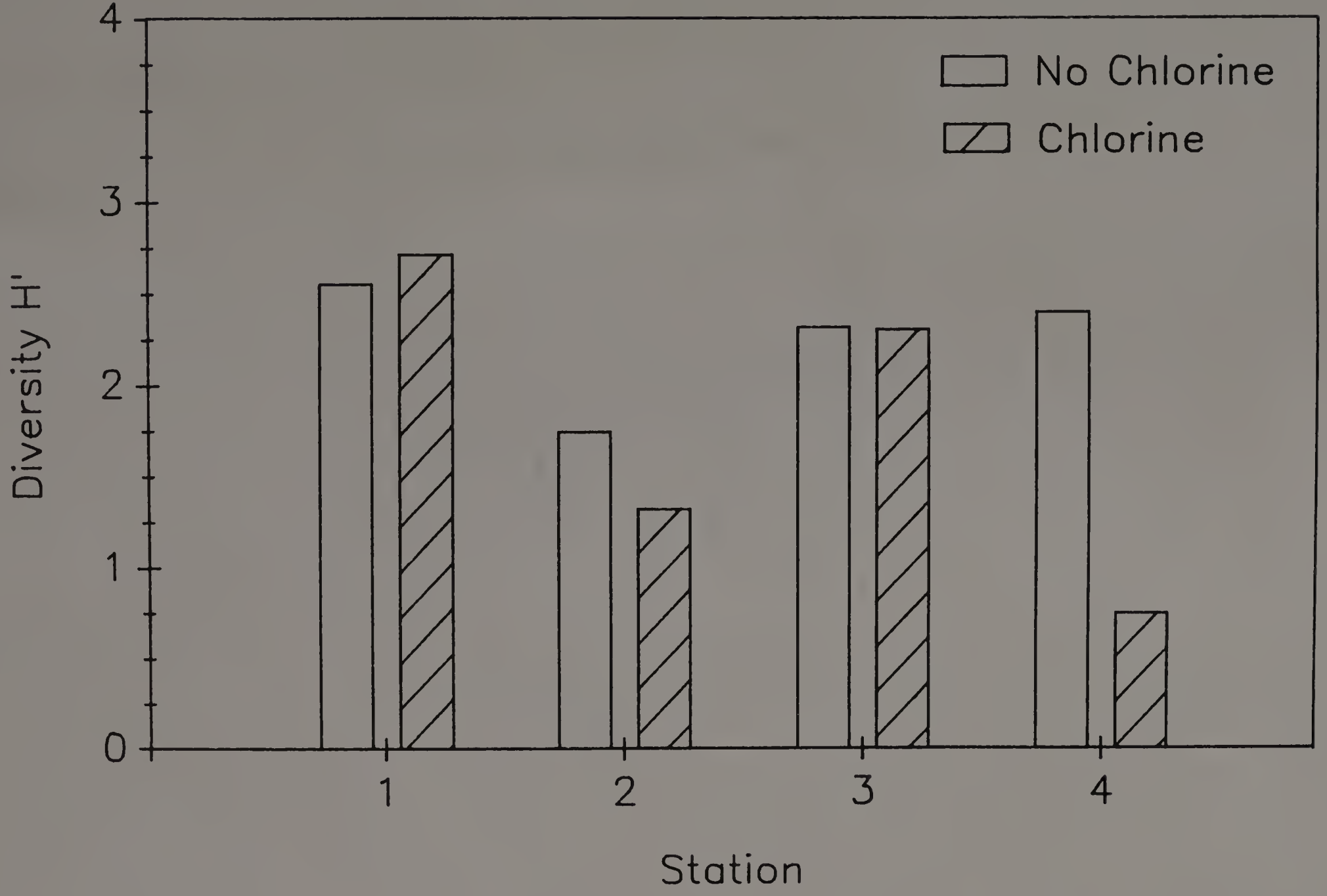

1
2
3
4
Diversity H'
0
1
2
3
4
−10
0
10
20
30
40
50
60
70
80
Time (Days Post−Chlorination)

Figure 7. Chironomidae diversity (Shannon-Wiener) at each station during the chlorination and post-chlorination periods. "No Chlorine" diversity based on pooled data from 20 samples collected on 5 January, 1990. "Chlorine" diversity based on pooled data from 11 samples collected on 27 October, 1989. Station 1 (control) is located 80 meters above the outfall and stations 2, 3, and 4 are located 80, 860, and 3700 meters below the outfall, respectively.


No Chlorine
Chlorine
Diversity H'
0
1
2
3
4
1
2
3
4
Station

CHAPTER 6
DISCUSSION

Because the effects of one stress can be modified greatly by another (i.e. sewage plus toxins) (Pagel and Langdon, 1981) it is useful, as a frame of reference, to explore what could generally be expected when only sewage is being discharged and how the addition of chlorine might change this. We can then see how well my data fits with these hypotheses.

Based on the high quality of the non-chlorinated wastewater entering Lampson Brook (Table A.3) and the latter's steep gradient one would not expect stress due to oxygen depletion during the study period. We could expect, however, enhanced primary and secondary productivity due to the organic enrichment at station 2. Further, at station 3 the residual wastewater combined with the inherent richness of a swampy region could be expected to result in a community of comparable magnitude, if not structure. After passing through a 500 hundred meter reach of turbulent flow, where the stream drops 25 feet in elevation, the allochthonous nutrient load could be expected to be reduced by the trickling filter effect in concert with molal action to control levels.

If it is assumed that TRC is the primary toxic fraction of the chlorinated sewage (Paller et al., 1988) and that the volatility and reactivity of TRC results in its short residency in well mixed streams (Reckhow, P.C.), then the station immediately downstream to the outfall (station 2) would experience the greatest depression in macroinvertebrate abundance. After 800 meters of mixing little if

any toxicity would be expected. Clearly, station 4 would not be exposed to any measurable levels of TRC and hence should not be impacted at all. Thus, only populations at station 2 should be reduced in abundance. A similar trend (i.e. The control being highest, station 2 the lowest, station 3 between station 2 and the control, and station 4 equal to the control.) would be expected for both diversity and taxa richness.

Chlorine, BOD, ammonia, dissolved oxygen, and phosphorous (Tables A.1 and A.2) are consistent with that expected for effluent and receiving water during the chlorinated and non-chlorinated periods. Further, after a 2 month period of non-chlorination (5 January, 1990) the total abundance data (Fig. 2) agreed remarkably well with the anticipated results. The diversity of the total community (Table 8), and the family Chironomidae (Fig. 7; Table 9) also supported this hypothesis, although the improvement at station four, relative to station 2, was not as great for H' as it was for total abundance (Table 8). The total number of different taxa was not consistent with the classical response to organic enrichment. station 4 had the least, stations 1 and 3 were about the same, and station 2 was between the control and station 4 (Table 8). The differences in the total number of Chironomid taxa were too small to reveal any statistical trends (Table 9). Although dependent upon sediment structure, the presence of Tubificidae (Table 5) is indicative of organic pollution (Brinkhurst, 1966; Aston, 1973). Also, Osbone (1987) found increased Oligocheates in the TRC plume of a municipal treatment plant. Thus their presence at stations 2 and 3 is consistent with the saprobien index.

The data from the chlorination period, however, point to an effect that is quite different from that expected. Instead of the abundance at station 2 being lower than when only sewage was discharged and stations 3 and 4 remaining unchanged, the opposite occurred. Station 2 was not significantly different from the non-chlorinated regime but stations 3 and 4 were significantly lower (Fig. 2; Table 4). Chironomidae abundance changed in a like manner with stations 3 and 4 decreasing. However, only station fours' change was significant (Fig. 4; Table 4). This observation indicates a degree of increased net toxicity of Belchertown's chlorinated wastewater with increasing distance from the outfall.

The differences in H' for total organisms at each station between chlorinated and 2 months post-chlorinated showed no notable trends, except for the increase at station 4 from 2.01 to 2.66 (Table 8). Chironomidae diversity, on the other hand, was unchanged at station 1 and 3, but was improved slightly at station 2 (1.33 to 1.75) and dramatically at station 4 (0.75 to 2.40) (Fig. 7; Table 8). As demonstrated by Perkins (1983), Shannons' H', being prone to false negatives but not false positives, is a conservative index. Consequently, it should be used qualitatively in conjunction with total abundance, taxa richness, and community similarity indices for the most reliable interpretations. It has even been suggested that taxa richness alone is a good indicator of stress (Dills and Rogers, 1974; Lenat, 1983). Taxa richness does in fact support the trends seen in total abundance. The total number of different taxa at station 4 doubled from 14 on 10 October, 1989 to

29 on 5 January, 1990. The average number of taxa per core increased significantly at station 4 (Table 10).

Although there is evidence for (DeSmett, 1982; Pratt et al., 1981) and against (Rosenberg and Weins, 1976; Lenat, 1983) the use of Chironomid taxa richness for the evaluation of water quality, here it was similar to taxa richness for the entire community. At station 4 total taxa richness increased 4 fold (Table 9) and average taxa richness increased significantly (0.75 to 5.73) with respect to the chlorination period (Fig. 5; Table 9).

However, Keefe at al. (1983) found slightly different trends. Their qualitative dip-net sampling, which they believed was the most consistent method between stations, yielded an increase in total organisms and total taxa at the furthest downstream station, relative to the control. The trends in their Surber samples, which they dismissed because of radical differences in sediment characteristics, were quite similar to our chlorinated regime. The mean density of the controls were remarkably low, the stations just below the outfall were approximately 10 fold greater than the control, and the station furthest from the control had the lowest mean density. On-site toxicity tests, in which dilutions of the wastewater effluent were evaluated, indicated that the postchlorinated/dechlorinated effluent was less toxic than the unchlorinated effluent. This was, however, a flowthrough test with no exposure to a sediment source. Arthur (1972) also found dechlorination removed toxicity. However, Esvelt (1973) noted a significant toxicity remained after dechlorination if the chlorine contact time lasted 3 days with a residual of 25% of the original

chlorine dose. This indicates the formation of a toxic fraction not found in Keefes' or Arthurs' effluents, or if present, not allowed to accumulate in a sediment sink/source for prolonged and increased exposure to the test animals.

The biological response observed in the chlorinated plume during the summer by Osborne and Davies (1987) is consistent with TRC being the toxic fraction (e.g. Increased abundance, decreased diversity, loss of Ephemeroptera and Chironomidae, and an increase in Oligocheta). Recovery at 1.5 Km was attributed to complete mixing at the first reversing meander (600 meters below the outfall), which diluted the TRC to levels below detection. However it is important to note that the average daily sewage discharge was between 0.1 % and 0.5 % of the average daily river discharge (i.e. a dilution factor of greater than 200 as compared to a dilution factor of between 2 and 5 for the present study) of the average daily discharge of the river. The high levels of TRC within 500 meters of the outfall were the result of a lack of mixing. Therefore the lack of an effect outside of the concentrated TRC plume may not be to due the absence of non-TRC factors but instead to the extreme dilution of any such factors. Furthermore, photosynthetic production would be high during their sampling period. This, combined with the limitation of toxicity to the narrow sewage plume, might lessen the the impact of chlorination on the food base of the macroinvertebrate community.

Stream levels of TRC at Lampson Brook have a limited persistence (Table A.2). Based on a laboratory determined half-life of 10 to 15 minutes for chloramines and a dye study on the 150

meters just below the outfall, Reckhow (P.C.) accurately predicted a residual of approximately 0.1 to 0.2 mg l^{-1} chloramines at 150 yards. As expected at station 3, nearly six times as far downstream, the TRC concentrations were rarely detectable (Table A.2). The high levels of D.O. preclude oxygen as a source of stress. Similarly, though ammonia was higher at Station 2 than the recommended level of 20 ug l^{-1} (EPA, 1986), stations 3 and 4 were not significantly different from the control or across time (Tables A.1 and A.2). Furthermore, Paller (1988) found that non-chlorinated secondary effluent had greater ammonia levels than chlorinated secondary but the impact was considerably less. Even with ammonia levels of 5 to 10 mg l^{-1}, the elimination of chlorination was more beneficial than the removal of the ammonia. This, coupled with the lack of any industrial wastes, suggests either the existence of some unidentified toxicant, which is either directly or indirectly related to the chlorination of the wastewater, or the downstream magnification of the biological rsponse to TRC toxicity.

The plot of total abundance against time (Fig. 1) shows station 2 decreased to control levels during the transition stages of acclimation to nonchlorinated effluent and then came back up on the final date. This could be partly due to the episodic rain events which occurred on 9 November, 1989 and 23 November, 1989 (Ives, 1989). Storms of this magnitude at a similar time of year resulted in an immediate stage increase of as much as ten inches. Although scouring would increase at all stations the increased depth and width would decrease the scouring effect at stations 3 and 4. Station 1, however, should also have been affected by this scouring.

Chemically or seasonally mediated shifts in the community are also likely factors. However, the continued dominance (Table 6) and similarity of the Chironomidae community on 11/10/89 and 11/24/89, as compared to the chlorination period (Table 13), results in uninterpretable fluctuations in the overall pre-equilibrium communities. Convoluted shifts in the overall community structure during the transition from the chlorinated regime to the non-chlorinated regime are also seen at station 4 (Tables 4 and 6).

The response of the Chironomidae community is not as erratic (Fig. 3; Table 4). The difference in significance between the total organism (Figs. 1 and 2) and Chironomidae abundances (Figs. 3 and 4) highlight some interesting community characteristics. Chironomidae was clearly less dominant at station 3 than at the other station (Table 6). Oligocheata were always prevalent at station 3 and were less prevalent at the control (Table 6). Furthermore, Crustacea, although present at all times, became dominant on the final sampling date. This shift in dominance from Chironomidae to Oligocheata and Crustacea (Table 6) resulted in a lack of significance in Chironomidae abundance between station 3 and the control on the first and final sampling dates (Fig. 4). Although the response of the entire indigenous macroinvertebrate community is of interest the absence of crustaceans at stations other than 3 may be due more to physical constraints than to chemical factors.

The inertia to change of the Chironomidae community on the first 2 sampling dates following cessation of chlorination (Fig. 3) is indicative of a long-term, severe perturbation and the absence of an

undisturbed source for recolonization (Cairns et al., 1971; Williams and Hynes, 1976). This is not observed in total abundance (Fig. 1) because of the increase in the abundance of Ephemeroptera-A on these dates (Table 4). This dramatic increase in very early instar mayflies causes the relative abundance figures for Chironomidae to fall even though their total number increased very slightly (Table 6). Furthermore, the greatest abundance of Ephemeroptera-A during chlorination is actually at station 2 (Table 4). Although Lenat (1983) preferred EPT taxa richness to Chironomid taxa richness, the use of Ephemeroptera-A as an indicator of pollution in the hyporheic zone may be tendentious. The lack of Ephemeroptera at station3 is most likely the result of the swampy environment surrounding the sampling station.

The non-significant drop in total abundance at station2 for the first 2 sampling dates during the post-chlorination is the opposite of what happened at the control and, to a greater degree, at station4 (Table 4). This temporary increase in early instar mayflies may be due to a hatch, which would account for the increased variability due to clumping (Resh, 1979), or the upward migration from the deeper hyporheic zone due to restricted pore space(Williams, 1984) or a seasonal migration (Williams and Hynes, 1976). Chironomidae may remain at greater depths because of their body shape or differences in tolerance levels (Williams, 1984). Whatever the cause by 5 January, 1990 they had returned to control levels. Hence it is possible that the Ephemeroptera-A experienced a fortuitous recruitment not undergone by Chironomidae. A residual toxicant

which affected the Chironomidae more than the mayflies may also have been a factor.

It could be argued that the observed changes are due to the interaction of seasonal changes and station specific differences, most important of which is the grain size (Cummins, 1962; Rabini and Minshall, 1977; Reice, 1980; Erman and Erman, 1984). This is not applicable to station 2. However, station 3 is downstream from swampy, poorly drained terrain, and station 4 is predominantly coarse sand and fine gravel, as opposed to the more heterogeneous sediments at the control (Table 1). The richer, siltier environment surrounding station 3 is better suited for oligochaetes and crustaceans than for mayflies (Cordone and Kelly, 1961) and, to a limited extent, for comparable Chironomidae communities. The sediment, however, did not change appreciably over time (Table 3) and there is no indication that Crustacea undergo a seasonal increase during the months in question. The larger grain size at station 4 may be selective for early instar Ephemeroptera-A but the removal of this taxa from the community only retards the return of total abundance to control levels. The subfamily Orthocladinae reportedly prefers coarser sediments (Pinder, 1980) but this taxa is a major component at the control station on all dates, including 5 January, 1990, when the sediment had become finer in composition (Tables 1 and 7). In fact, on the final sampling date Orthocladinae were more prevalent at the control station than Chironominae and Tanypodinae (Table 7), which prefer finer sediments (Pinder, 1980). In summary, the observed site specific differences do not explain the changes in community structure.

The use of Chironomidae to classify lakes and streams has focused almost exclusively on the correlation of community composition with nutrient enrichment and saprobiety (Learner et al, 1971; Wilson and Bright, 1973; Brinkhurst, 1974; Beck, 1977; Saether, 1979; Kawai et al., 1989). However, biotic indices are limited to specific types of pollution and the geographic region in which the tolerances were developed (Washington, 1984). Armitage and Blackburn (1985) attempted to use tolerance codes based on organic enrichment and "toxic pollutants" (Willson and McGill, 1982) to classify the impact of zinc and organic enrichment on lotic Chironomidae communities. Six of the twelve genera from the most polluted stations were classified as intolerant to relatively intolerant.

The response of the Chironomidae community to specific heavy metal (Surber, 1957; Wentsel et al., 1977; Winner et al., 1980; Armitage and Blackburn, 1985) and petroleum pollution (Tubb and Dorris, 1965; Rosenberg and Wiens, 1976) have been documented. However, the paucity of research regarding chironomid community alterations due to chlorinated compounds (Pinder, 1986) prevents the clears interpretation of the changes in this community. Although the development of such an index is beyond the scope of this investigation, some of the genera exhibited trends which show promise for the future exploration of the effects of chlorinated sewage and thus warrant speculation.

The high percentage of Chironominae at station 2 is indicative of organic pollution (Armitage and Blackburn, 1985). Kawai et al. (1989) observed a negative correlation between B.O.D. and

Chironomid diversity. Polypedilum was frequently found in organically enriched streams. This is consistent with the increased B.O.D. and Polypedilum at station 2 (Tables A.2 and 5). However, Beck (1977) classified Polypedilum fallax as saprophobic to facultative and Polypedilum illinoense as saproxenous to facultative. Unfortunately the lack of consensus in the literature reviewed by Beck (1977) obfuscates any relationships which may be present.

Paratendipes was more prevalent at station 2 than the control on all dates except 5 January, 1990 (Table 5). The temporal reduction in this genera contributed to the shift in dominance from Chironominae to Chironominae and Orthocladinae at station 2 (Table 7). This change could be naturally or chemically mediated. Unfortunately, even if species level identifications had been made, life history could not be easily excluded due to the variability in the literature at hand (Beck,1977). The increase in Micropsectra at station 3 only is a clear indication of habitat preference. Although the life cycle varies even within 1 species (Beck, 1977) it is possible that this increase is chemically mediated. The presence of Heleniella on all dates but only at the control is quite interesting (Table 5). Its absence at station 4 on 5 January, 1990 makes interpretation difficult. The lack of its inclusion in The New England Macroinvertebrate Checklist (Bilger, 1986) could be indicative of a very specialized population. Hydrobaenous was only present at the control and station 3 during chlorination. By 5 January, 1990 it had become significantly more abundant at station 4 than at the control (Table 5). This genus may be a useful marker for the effects observed in this study. Why it rose above control levels is unclear

but its absence during chlorination may be due to a selective stressor. Once again the limited natural history for this genus, coupled with the unkown nature of the stressor, permits speculation only. Clearly there are some interesting possibilities for future research here. The first step should be towards the clear identification of the source of the stress at station 4.

The elimination of measured chemical or natural causes of the observed effects of chlorinated wastewaters leads to speculation on the possible existence of unmeasured toxicants or biological interactions.

Based on the concurrence of toxicity and chlorination the most likely toxicant is a chlorinated by-product or combination of by-products. Many researchers have found increased levels of a variety of chlorinated organic compounds in domestic wastewater (Jolley, 1975; Murphy, 1975; Kintsley et al., 1983; Trehey et al., 1986; Oliver and Visser, 1980; Miller and Uden, 1983; Boyce and Hornig, 1983) and bleached pulp and papermill effluents (Paasivirta, 1988). These chlorinated organic compounds have been shown to be toxic. Furthermore, chlorinated organic compounds, especially non-polar compounds, can exist in the sediments for a long time (Larsson, 1985 and 1986).

Moore et al.(1980) and Osborne (1982) observed an increase in mutagenic activity in fish, benthic macroinvertebrates, and concentrated (100x) water samples within 100 meters of a chlorinated municipal sewage outfall. The authors, however, observed no mutagenic activity in the sediments and postulated that the mutagenic activity in the macroinvertebrates was not due to

direct accumulation from the food source. However they did conclude that the mutagenic activity would be expected to increase along the aquatic food chain.

Schuytema (1988) found the sediment to be an important exposure route of hexachlorobenzene for the fathead minnow and, at times, for lumbriculus. Although Adams et al. (1985) did not determine the sediments to be an important uptake route of kepone in Chironomus tentans he did find the interstitial water and the water at the sediment surface to be important sources. Similarly, Edie et al. (1982) speculated on the importance of pore water as a source of PAH for the Amphipod Pontoporeia hoyi. Reckhow (P.C.) found increased levels of halogenated organics (TOX) in the sediment pore water at Lampson Brook. They also observed an increase in pore water TOX with increasing distance from the outfall and speculated on the instream formation of TOX. This results in an increase in chlorine contact time with a variety of organic compounds. Furthermore, Esvelt (1973) found that increased chlorine contact time resulted in increased toxicity of dechlorinated sewage effluent. Unfortunately Reckhows' sampling area only extended 150 meters downstream from the outfall.

Deploying baskets of 2.5 cm. diameter stones as artificial substrates in the water column, Coler (1990) documented similar effects of Belchertown's sewage discharge on the macroinvertebrates of Lampson Brook. Although collected at the end of the chlorination season the difference in substrate characteristics (i.e. Approximately 2.5 inch diameter stones suspended in the water column) prevents a direct comparison to the

present investigation. Even so, many of the genera of Chironomidae identified in the cores (i.e. Diplocladius, Hydrobaenous, Micropsectra, and Polypedilum.) were present in her substrates (Coler, 1990). The loss of sensitive taxa and increased total abundance at stations 2 and 3 was indicative of organic enrichment and TRC toxicity. Although the sensitive taxa returned at 3700 meters below the outfall the recovery was incomplete when compared to the control. It was postulated that the residual toxicity at 3700 meters may have been due to a sediment-borne toxicant. In general, the previous findings at Lampson Brook are supportive of those observed presently.

A biological explaination for the current observations might be linked to litter processing. Headwater streams (first to third order) are primarily heterotrophic with leaf litter being a principle component of the allochthanous energy (Fisher and Likens, 1973; Cummins, 1974; Anderson and Sedell, 1979; Cummins and Klug, 1979; Minshall et al., 1983). In fact, the growth of winter-active macroinvertebrates is dependant upon the seasonal infux of allochthanous material (Hynes, 1961). The autumnal increase in leaf litter quality is a direct benefit for shredders. Collectors and gatherers benefit from the processing of coarse particulate organic matter (CPOM > 1 mm.) into fine particulate organic matter (FPOM < 1 mm.) (Anderson and Sedell, 1979). Even grazers and predators may be detitivores in their earliest stages of development (Coffman et al., 1971).

It has been documented (Wallace et al., 1970; Barlocher and Kendrick, 1973a and 1973b) that the microbial flora colonizing CPOM

has a greater nutritional value than the leaf itself, which is mostly cellulose and lignins. The importance of the microbial flora, which is mostly fungi (Suberkropp and Klug, 1976), is underscored by the preference of shredders for micobialy conditioned material (Kaushik and Hynes, 1971; Iversen, 1973; Barlocher and Kendrick, 1975a and 1975b). The contribution of the micobial biomass to shredders may be due to their inherent nutritional value (i.e. the "peanut butter" on the "cracker" Cummins, 1974)) or, indirectly, to an increase in the nutritional value of the partialy transformed substrate (Barlocher and Kendrick, 1975b).

A decrease in litter processing due to reduced microbial microbial colonization would result in reduced quality and/or quantity of FPOM. Consequently, collectors and gatherers downstream would be adversly affected.

Reduction of shredders by direct toxicity of TRC would also reduce the downstream quantity of FPOM. Newman et al. (1987) observed a decrease in litter processing in outdoor experimental stream exposed to 230 ug l^{-1} TRC. The authors believed both initial microbial conditioning and shredder colonization to be important factors. Shredder colonization, especially by the amphipods Crangonyx sp. and Hyalella azteca (Saussure), was considered more important than microbial colonization.

Since densities of both amphipods and chironomids, which are primarily gatherers and collectors (Merritt and Cummins, 1984), were reduced downstream from the Belchertown discharge, it is conceivable that decreased quality and quantity of food was a factor. Therefore two modes of action of TRC may be responsible for the

observed impact of chlorinated sewage at 80 and 3700 meters below the outfall. The lower diversity at station 2, just below the outfall, may be due to the presence of TRC. The decrease in diversity and abundance at station 4 may be a result of the abacence of a high quality food source. Microbially conditioned CPOM and FPOM may be transported into station 2 from above the sewage outfall. However, the CPOM and FPOM being transported down to station 4 would have been exposed to TRC during the colonization period. This could reduce both the quality and quantity of detritus at station 4.

In general, it appears that chlorination of Belchertowns' wastewater effluent may affect Lampson Brook through three modes of action; direct toxicity of TRC, indirect toxicity through the formation of TOX, and indirect toxicity through the disruption of the food chain. One or all avenues of impact may be occuring. This investigation was designed to ascertain wether or not the effect of chlorinated wastewater, from Belchertown Wastwater Treatment Plant, is different than the effect of non-chlorinated wastewater. Identification of the mode or modes of action is beyond the scope of this study. Furthermore, the lack of true stream system replication prohibits the extrapolation of these results to chlorinated wastewater effluents in general.

CHAPTER 7

CONCLUSIONS AND RECOMMENDATIONS

7.1 Conclusions

Several conclusions regarding the effects of chlorinated sewage on the macroinvertebrates of Lampson Brook can be drawn from this research.

1) Non-chlorinated sewage enriched stations two and three, as demonstrated by the increase in abundance, but had little effect on station four.

2) All treatment stations were affected by chlorinated sewage. TRC was the toxic factor just below the outfall. The greatest impact with regards to reductions in density and diversity occurred at station four, 3700 meters below the sewage discharge, and was not due to TRC, oxygen depletion, or ammonia.

3) The toxic fraction may be sediment bound, as evinced by the greater response of the hyporheos, compared to the organisms colonizing substrates suspended above the river bed.

4) The effect may be food chain related, based on the downstream magnification of the impact.

7.2 Recommendations

Several recommendations for further research on this phenomena follow:

1) Multi-seasonal sampling with more intermediate treatment stations.

2) Increased control and indentification of variables through the measurement of TOX, the development of hyporheic artificial substrates, measurement of the affect on the downstream food base, and laboratory verification of results.

3) Correlation of Chironomidae with the toxic fraction of chlorinated wastewater for the development of a biotic index.

4) Investigation of other streams in order to determine wether or not this is a localized phenomenon.

APPENDIX

CHEMICAL DATA

Table A.1. Average (± 1.96 * SE), minimum, and maximum concentration of chemical parameters from the 1988 non-chlorination season (February and March). All measurements are in mg l^{-1} except pH and temperature (degrees centigrade).

Parameter	Statistic	Station 1	Station 2	Station 3	Station 4
Acidity	N	5	5	5	5
	Ave ± 1.96*SE	3.5 ± 0.3	4.9 ± 0.3	8.5 ± 2.0	4.7 ± 1.2
	(Min , Max.)	(2.9 , 3.8)	(4.5 , 5.4)	(5.9 , 12.1)	(3.8 , 7.0)
Alkalinity	N	5	5	5	5
	Ave ± 1.96*SE	30.8 ± 3.2	33.9 ± 2.1	27.9 ± 2.8	22.2 ± 1.7
	(Min , Max)	(25.2 , 34.4)	(31.5 , 36.8)	(25.2 , 32.8)	(20.0 , 24.2)
Ammonia	N	5	5	5	5
	Ave ± 1.96*SE	0.20 ± 0.10	0.40 ± 0.32	0.30 ± 0.21	0.29 ± 0.18
	(Min , Max)	(0.09 , 0.34)	(0.11 , 0.98)	(0.00 , 0.58)	(0.00 , 0.51)
B.O.D.	N	5	5	5	5
	Ave ± 1.96*SE	1.7 ± 0.7	4.9 ± 3.3	1.9 ± 0.5	1.6 ± 0.7
	(Min , Max)	(0.4 , 2.6)	(2.5 , 11.7)	(1.2 , 2.5)	(0.6 , 2.8)
Calcium	N	5	5	5	5
	Ave ± 1.96*SE	13.7 ± 0.9	14.6 ± 1.0	13.0 ± 0.9	9.1 ± 2.0
	(Min , Max)	(12.1 , 14.8)	(12.8 , 15.6)	(11.4 , 14.0)	(5.5 , 11.4)

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Table A.1 (Continued).

Parameter	Statistic	Station 1	Station 2	Station 3	Station 4
Chloride	N	5	5	5	5
	Ave ± 1.96*SE	29.4 ± 5.3	35.6 ± 5.2	29.2 ± 4.5	25.7 ± 6.9
	(Min , Max)	(21.7 , 37.5)	(27.9 , 42.4)	(24.3 , 36.4)	(17.0 , 36.5)
D.O.	N	5	5	5	5
	Ave ± 1.96*SE	11.4 ± 0.6	11.0 ± 1.0	10.9 ± 0.4	11.6 ± 0.4
	(Min , Max)	(10.3 , 11.9)	(10.1 , 11.6)	(10.5 , 11.6)	(11.0 , 12.1)
Hardness	N	5	5	5	4
	Ave ± 1.96*SE	62.4 ± 6.3	61.5 ± 4.5	54.5 ± 3.8	46.7 ± 3.1
	(Min , Max)	(54.0 , 71.6)	(57.2 , 69.6)	(50.0 , 59.6)	(43.6 , 50.4)
Nitrate	N	5	5	5	5
	Ave ± 1.96*SE	0.36 ± 0.05	0.55 ± 0.09	0.45 ± 0.07	0.39 ± 0.04
	(Min , Max)	(0.27 , 0.44)	(0.46 , 0.68)	(0.35 ,0.56)	(0.32 , 0.45)
pH	N	5	5	5	4
	Ave ± 1.96*SE	7.3 ± 0.1	7.3 ± 0.1	7.0 ± 0.1	7.1 ± 0.1
	(Min , Max)	(7.1 , 7.40)	(7.2 , 7.4)	(6.9 , 7.1)	(6.9 , 7.2)
Phosphorous	N	5	5	5	5
	Ave ± 1.96*SE	0.09 ± 0.06	0.43 ± 0.08	0.31 ± 0.09	0.14 ± 0.03
	(Min , Max)	(0.03 , 0.17)	(0.30 , 0.53)	(0.23 , 0.49)	(0.10 , 0.18)

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Table A.1 (Continued).

Parameter	Statistic	Station 1	Station 2	Station 3	Station 4
Suspended Solids	N	5	5	5	5
	Ave ± 1.96*SE	10 ± 9	10 ± 7	3 ± 2	5 ± 4
	(Min , Max)	(2 , 29)	(4 , 20)	(1 , 6)	(1 , 13)
Total Solids	N	5	5	5	5
	Ave ± 1.96*SE	167 ± 77	165 ± 42	127 ± 44	108 ± 43
	(Min , Max)	(104 , 319)	(97 , 224)	(89 , 212)	(63 , 188)
Temperature	N	3	4	4	4
	Ave ± 1.96*SE	4.0 ± 1.1	5.9 ± 2.4	5.1 ± 3.1	4.1 ± 2.3
	(Min , Max)	(3.0 , 5.0)	(4.0 , 9.5)	(2.0 , 9.5)	(1.0 , 6.8)
TRC	N	0	0	0	0
	Ave ± 1.96*SE	N/A	N/A	N/A	N/A
	(Min , Max)	N/A	N/A	N/A	N/A

N = Number samples.
N/A = Not analyzed.

Table A.2 Average (± 1.96 * SE), minimum, and maximum concentration of chemical parameters from the 1988 chlorination season (October). All measurements in are mg L^{-1} except pH and temperature (degrees centigrade).

Parameter	Statistic	Station 1	2	3	4
Acidity	N	7	7	7	7
	Ave ± 1.96*SE	3.9 ± 0.9	5.0 ± 1.5	9.0 ± 2.7	4.6 ± 1.2
	(Min , Max.)	(2.4 , 6.0)	(3.2 , 8.0)	(4.8 , 15.9)	(2.6 , 6.8)
Alkalinity	N	7	7	7	7
	Ave ± 1.96*SE	41.1 ± 4.9	44.3 ± 3.4	39.9 ± 3.6	31.9 ± 3.0
	(Min , Max.)	(30.0 , 49.4)	(38.1 , 52.4)	(33.0 , 46.4)	(27.0 , 37.1)
Ammonia	N	6	6	6	6
	Ave ± 1.96*SE	0.30 ± 0.19	0.59 ± 0.27	0.28 ± 0.15	0.25 ± 0.13
	(Min , Max.)	(0.06 , 0.61)	(0.10 , 0.98)	(0.09 , 0.48)	(0.00 , 0.46)
B.O.D.	N	7	7	7	7
	Ave ± 1.96*SE	2.9 ± 1.1	5.4 ± 2.2	2.3 ± 1.2	2.4 ± 1.0
	(Min , Max.)	(1.1 , 4.9)	(1.5 , 9.7)	(0.9 , 5.5)	(1.3 , 5.2)
Calcium	N	7	7	7	6
	Ave ± 1.96*SE	23.9 ± 3.2	23.1 ± 5.4	21.1 ± 4.6	16.8 ± 3.1
	(Min , Max.)	(19.5 , 32.7)	(18.4 , 39.3)	(18.0 , 35.0)	(13.8 , 24.5)

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Table A.2 (Continued).

Parameter	Statistic	Station 1	Station 2	Station 3	Station 4
Chlorides	N	7	7	7	7
	Ave ± 1.96*SE	40.8 ± 5.1	47.8 ± 8.4	43.9 ± 9.0	32.2 ± 6.7
	(Min , Max.)	(32.7 , 50.0)	(38.8 , 70.7)	(35.0 , 68.5)	(25.0 , 51.8)
D.O.	N	7	7	7	7
	Ave ± 1.96*SE	9.7 ± 0.8	8.8 ± 0.8	6.3 ± 0.7	9.8 ± 0.8
	(Min , Max.)	(8.3 , 11.3)	(7.4 , 10.4)	(5.3 , 7.9)	(8.4 , 11.5)
Hardness	N	7	7	7	7
	Ave ± 1.96*SE	82.9 ± 3.0	80.3 ± 3.8	75.3 ± 3.3	63.0 ± 4.2
	(Min , Max.)	(76.4 , 86.7)	(76.0 , 88.4)	(71.2 , 83.2)	(56.0 , 72.0)
Nitrate	N	7	7	7	7
	Ave ± 1.96*SE	0.24 ± 0.35	0.34 ± 0.10	0.36 ± 0.22	0.07 ± 0.04
	(Min , Max.)	(0.01 , 1.30)	(0.20 , 0.57)	(0.12 , 0.83)	(0.02 , 0.15)
pH	N	7	7	7	7
	Ave ± 1.96*SE	7.1 ± 0.2	7.1 ± 0.2	6.8 ± 0.3	7.1 ± 0.3
	(Min , Max.)	(6.8 , 7.5)	(7.05 ,7.5)	(6.5 , 7.4)	(6.5 , 7.5)
Phosphorous	N	7	7	7	7
	Ave ± 1.96*SE	0.09 ± 0.05	1.04 ± 0.23	0.67 ± 0.25	0.27 ± 0.06
	(Min , Max.)	(0.04 , 0.22)	(0.81 , 1.72)	(0.43 , 1.29)	(0.18 , 0.40)

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Table A.2 (Continued).

Parameter	Statistic	Station 1	Station 2	Station 3	Station 4
Suspended Solids	N	7	7	7	7
	Ave ± 1.96*SE	1 ± 1	3 ± 2	1 ± 1	2 ± 1
	(Min , Max.)	(1 , 3)	(1 , 9)	(1 , 3)	(1 , 5)
Total Solids	N	7	7	7	7
	Ave ± 1.96*SE	188 ± 35	229 ± 30	204 ± 25	162 ± 39
	(Min , Max.)	(147 , 286)	(185 , 309)	(146 , 301)	(106 , 266)
Temperature	N	7	7	7	7
	Ave ± 1.96*SE	10.5 ± 2.3	12.3 ± 1.9	10.6 ± 2.1	10.6 ± 1.8
	(Min , Max.)	(7.0 , 16.0)	(9.0 , 16.5)	(7.5 , 15.0)	(7.0 , 15.2)
TRC*	N	10	9	10	10
	Ave ± 1.96*SE	0.00 ± 0.00	0.47 ± 0.15	0.01 ± .01	0.00 ± 0.00
	(Min , Max.)	(0.00 , 0.00)	(0.24 , 0.82)	(0.00 , 0.03)	(0.00 , 0.00)

N = Number samples.

* Includes samples from April, 1988.

Table A.3 Average (± 1.96 * SE), minimum, and maximum concentration of chemical parameters of the Belchertown wastwater effluent from the 1988 non-chlorination season (February and March) and the chlorination season (October). All measurements are in mg L^{-1} except pH and temperature (degrees centigrade).

		Period	
Parameter	Statistic	Non-Chlorination	Chlorination
Acidity	N	5	7
	Ave ± 1.96*SE	10.8 ± 3.2	9.4 ± 3.2
	(Min , Max)	(4.6 , 13.2)	(4.8 , 17.6)
Alkalinity	N	5	7
	Ave ± 1.96*SE	58.4 ± 3.6	46.9 ± 6.8
	(Min , Max)	(53.6 , 64.1)	(40.0 , 61.7)
Ammonia	N	5	6
	Ave ± 1.96*SE	2.47 ± 0.93	1.39 ± 0.59
	(Min , Max)	(1.71 , 4.27)	(0.00 , 0.46)
B.O.D.	N	5	6
	Ave ± 1.96*SE	5.1 ± 0.6	5.6 ± 1.9
	(Min , Max)	(3.9 , 5.6)	(3.4 , 9.7)
Calcium	N	5	7
	Ave ± 1.96*SE	17.1 ± 0.7	23.4 ± 11.1
	(Min , Max)	(15.8 , 17.8)	(15.2 , 56.9)
Chloride	N	5	7
	Ave ± 1.96*SE	59.8 ± 2.5	59.5 ± 4.6
	(Min , Max)	(56.5 , 63.1)	(55.5 , 73.5)
D.O.	N	5	7
	Ave ± 1.96*SE	9.8 ± 0.6	8.0 ± 0.8
	(Min , Max)	(8.6 , 10.4)	(3.4 , 9.7)
Hardness	N	5	7
	Ave ± 1.96*SE	74.2 ± 1.7	69.2 ± 3.9
	(Min , Max)	(71.6 , 76.4)	(61.2 , 76.4)

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Table A.3 (Continued).

Parameter	Statistic	Period	
		Non-Chlorination	Chlorination
Nitrate	N	5	7
	Ave ± 1.96*SE	1.07 ± 0.54	0.68 ± 0.24
	(Min , Max)	(0.56 , 2.13)	(0.29 , 1.17)
pH	N	5	7
	Ave ± 1.96*SE	7.2 ± 0.1	6.9 ± 0.2
	(Min , Max)	(7.1 , 7.2)	(6.7 , 7.4)
Phosphorous	N	5	7
	Ave ± 1.96*SE	1.80 ± 0.18	2.25 ± 0.34
	(Min , Max)	(1.55 , 2.00)	(1.9 , 3.3)
Suspended Solids	N	5	7
	Ave ± 1.96*SE	7 ± 5	4 ± 3
	(Min , Max)	(3 , 16)	(1 , 13)
Total Solids	N	5	7
	Ave ± 1.96*SE	227 ± 31	288 ± 28
	(Min , Max)	(192 , 286)	(253 , 349)
Temperature	N	3	7
	Ave ± 1.96*SE	6.5 ± 1.5	13.0 ± 2.4
	(Min , Max)	(5.0 , 7.5)	(9.5 , 18.0)
TRC*	N	0	10
	Ave ± 1.96*SE	N/A	1.91 ± 0.61
	(Min , Max)	N/A	(0.93 , 3.29)

N = Number of samples.
N/A = Not analyzed.
* Includes samples from April, 1988.

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