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**TRANSPORT AND FATE
OF MICROBIAL CONTAMINANTS
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IN THE GREAT BAY: EFFECTS ON
WATER QUALITY AND MANAGEMENT
IMPLICATIONS**

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TRANSPORT AND FATE OF MICROBIAL CONTAMINANTS AND SUSPENDED SEDIMENTS IN THE GREAT BAY: EFFECTS ON WATER QUALITY AND MANAGEMENT IMPLICATIONS

INTRODUCTION

The incidence of waterborne disease in the United States has been on the increase for over two decades (1). Many diseases are the result of the fecal-oral route of disease transmission that is often associated with direct consumption or contact with contaminated water. Sewage and other sources of fecal microorganisms that may be discharged into estuarine waters pose a public health hazard for all recreational users, but the most acute potential effect is on shellfish consumers. Shellfish are the estuarine resource most often affected by pollution. Shellfish harvesting in estuarine waters throughout the United States is often limited because concentrations of standard fecal indicator bacteria exceed safe limits. In many places, significant efforts have been made to eliminate major point sources of fecal contamination in estuarine waters. In New Hampshire, \$60 million has been spent to upgrade publicly owned wastewater treatment works (POTW) in coastal communities (2). At present, all of the communities surrounding the Great Bay estuary (Figure 1) have upgraded POTWs, which should diminish their importance as sources of microbial contamination. However, virtually nothing is known about nonpoint source pollution in coastal New Hampshire. This remains a problem in coastal New Hampshire, and much public attention has recently been directed to the closing of shellfish beds because of rising levels of fecal-borne microbial contaminants in shellfish growing waters (3).

Seven major rivers empty into the Great Bay estuary. Many of these rivers are also the receiving waters for municipal wastewater discharges for communities located on the rivers. Recent research and monitoring studies conducted at JEL have been directed to the study of

bacterial pathogens and indicators of fecal pollution, including total and fecal coliforms, Escherichia coli, and enterococci, in estuarine water and shellfish (4-7). One of the most striking trends determined from these studies is the consistent incidence of higher levels of indicators at the Furber Strait area of Adams Point between Little and Great Bays at high tide than at low tide. This evidence and the relatively low levels of bacterial contamination in the Great Bay, despite higher levels in all surrounding waters, suggest that the Bay may be a sink for contaminants. If the natural processes causing the removal of microorganisms could be determined, then methods could be developed to help control water pollution problems. Such strategies would be relatively low-cost alternative management practices for controlling pollutant inputs into Great Bay.

A number of different types of research studies have been conducted that relate to the present study. For example, Officer et al. (8) attributed the paucity of phytoplankton blooms in San Francisco Bay to the activities of the filter feeding benthic community. The control of phytoplankton and nutrient levels was attributed to favorable environmental factors and the activities of the high numbers of clams and mussels in the bay. Ward et al. (9) conducted studies on the impact of seagrasses on suspended sediments in Chesapeake Bay. This work showed seagrass dramatically reduced suspended sediment concentrations and enhanced deposition within the beds. Thus, natural processes in different bays have been shown to have favorable impacts on water quality.

Implicit in the present study is a relationship between suspended solids and bacteria, particularly indicators of fecal contamination. Plummer et al. (10) found that most bacterial activity in turbid estuarine waters was associated with bacteria attached to particles that remain almost permanently in suspension (PSP), which have relatively large mean sizes and organic content. They found, as others have shown (11), that resuspended sediments stimulate the activities of both attached and free-living bacteria. However, PSP particles were found to be much more densely colonized than resuspended particles, suggesting that different types of suspended particles may have different types of associations with bacterial populations. In addition, Albright et al. (12)

showed increases in percent attached bacteria were inversely proportional to chlorophyll a content and primary productivities, and were not related to salinity or silt levels. Attachment of bacteria to particles was thus related to the physiological condition of phytoplankton, their major carbon and energy sources: bacteria tended to attach to senescent phytoplankton and become free-living during periods of phytoplankton growth. Thus, bacterial attachment and association with suspended particles depends on environmental factors and the source and nature of the particles.

The ability for shellfish to filter suspended particles from water is well known. In relation to the sanitary quality of freshly-harvested shellfish, this activity is both the root of the problem and the best strategy for eliminating the problem. Shellfish filter, accumulate, and retain microbial pathogens for varying lengths of time depending on the conditions of exposure (13). Conversely, exposure of shellfish to relatively clean or uncontaminated water through the processes of relaying and depuration have been shown to be effective strategies for enhancing elimination of pathogens from shellfish (7,14). Oysters and especially mussels have been shown to acquire significant levels of their metabolic carbon and nitrogen requirements from filtering suspended bacteria (15). In contrast, Wright et al. (16) found mussel beds in the field were effective in removing phytoplankton but not bacteria. However, Albright et al. (12) showed bacterial attachment to phytoplankton to be significant under certain circumstances, suggesting that removal of bacteria with the phytoplankton could also have occurred in the Wright et al. (16) study. Thus, suspended bacteria can be taken up, retained, and/or eliminated from shellfish, depending on the activity of the shellfish and the nature of any particle association.

Eelgrass leaves form a three-dimensional baffle in estuarine waters that acts as a damper to reduce water motion. Eelgrass meadows may filter estuarine waters, removing both suspended sediments and dissolved nutrients (17). Suspended materials carried by currents move into eelgrass beds and are rapidly settled to the bottom. Epiphytic growth is often stimulated in areas of poor water quality (18). The blades and the epiphytes appear dull brown coated with a fine layer of sediment, and they often sink to the bottom. When eelgrass in this condition is disturbed, a

cloud of sediment is released into the water. The magnitude of smothering by sediments in an estuarine environment is difficult to predict, although it has the potential for eliminating eelgrass from heavily impacted areas.

The overall goal of the study was to determine the causes of observed reductions of microbial contaminants in Great Bay, so that better management practices can be applied throughout the estuary and surrounding watersheds to promote mitigation of microbial water quality problems. The specific objectives of this study were to identify processes by which microbial contaminants are lowered as a result of transport through the estuary; to demonstrate removal of microbial contaminants as water moves across eelgrass and shellfish beds; and to characterize the relationship between suspended solids and microbial concentrations.

The focus of this study was on the potential for natural processes, probably associated with shellfish and eelgrass beds, to act as natural filters that remove suspended microbial contaminants from the water. Eelgrass and shellfish beds are common to Great Bay and other temperate estuaries, yet their potential role in removing pollutants has not been fully investigated. Since at least 1960 (19), some portion of Great Bay has been open for shellfishing, based on the incidence of relatively low levels of microbial contaminants. Thus, the water appears to be cleaner in terms of coliform bacteria despite the presence of higher concentrations of contaminants in all other areas of the surrounding estuarine waters.

METHODOLOGY

Synoptic surveys of target microorganisms, total suspended sediments (TSS) and general water quality characteristics (salinity, temperature, etc.) were conducted at sites within the Great Bay to determine the distribution of microbial and TSS levels as water flowed across the Bay. Hand-collected surface samples were collected at three sites (Figure 1) approximately every two weeks at low and high tides during summer, fall and spring. Intensified sampling took place

during the week following Hurricane Bob during August, 1991. A sampling transect across Great Bay was established with stations located at the entrance to Great Bay off Adams Point at mid-channel (AP), at a mid-bay location where the main channels meet (MB=Mid Bay), and at the southern end of the Bay at a mid-channel site within the Squamscott River off Chapmans Landing (SR). In total, surface samples were collected 31 times from June 30, 1991 to June 9, 1992. This sampling allowed for evaluation of the major source of microbial contaminants to the Bay at SR, concentrations leaving the Bay with tidal flow at AP, and concentrations at the center of the Bay (MB) that should reflect contaminant removal processes. Water temperature, conductivity, and salinity were measured using a Yellow Springs CTS meter.

More detailed assessment of the general distribution of TSS concentrations in the Great Bay were determined. Sampling cruises were conducted at 4 to 15 day intervals from August 13, 1991 to October 16, 1991, once in mid-November, 1991, at 10 to 16 day intervals from April 29, 1992 to August 18, 1992, and then at 3 to 4 week intervals from September to the end of October, 1992. In total, 20 cruises were conducted during this period during which water samples were collected (with a submersible pump) normally at all three stations and at three depths (surface, mid-depth and 1 m off the bottom). On a few occasions, we were not able to occupy the MB station or we had to sample from the docks at Jackson Estuarine Laboratory and under the Route 108 Bridge (rather than at the mid-channel sites at AP or SR). Sampling was conducted within 1 or 2 hours of low tide during all cruises and within 2 hours of high tide during 14 cruises; samples were collected only during low water on 6 occasions. In this report, only the samples collected from August, 1991 to June, 1992 are discussed.

The microorganisms detected were total (TC) and fecal (FC) coliforms, enterococci, E. coli (Ec) and Clostridium perfringens (Cp). Samples were collected as surface grabs in sterile 1 L plastic bottles. Enterococci, the standard indicator currently recommended by the EPA for estuarine waters, was the microbial indicator that was used most consistently, while C. perfringens was added to the study during the spring and became increasingly important as the study

progressed. Enterococci were measured using a standard membrane filtration method and mE agar with standard confirmation steps (20). The coliforms and E. coli were measured using a standard multiple tube fermentation, MPN analysis (21) that incorporates a fluorogenic molecule, 4-methylumbelliferyl-B-D-glucuronide (MUG) into the FC confirmed medium (EC) for E. coli determinations (22). C. perfringens, which is a conservative tracer of fecally-contaminated water, was detected using membrane filtration and mCP agar (23). The presence of enterotoxigenic E. coli using an adapted polymerase chain reaction method was not possible, as the method was not developed to the extent that it could be used for environmental samples.

The water samples were analyzed for TSS and archived for future analysis of organic content [combustible fraction or particulate organic carbon (POC) and particulate organic nitrogen (PON)]. Samples for TSS determinations were placed in 1 L plastic storage bottles and stored in the dark on ice until analyzed in the laboratory (within 24 hours). TSS were determined gravimetrically by filtering approximately 500 to 1000 ml water samples through 47 mm diameter, 0.3 micron nominal pore diameter prewashed, preweighed glass fiber filters. The filters were dried at less than 50° C for 24 hours for determination of total weight. All samples were combusted at 450°C for 4 hours to determine approximate organic content. For those samples selected for POC and PON analysis, a 50 ml aliquot was field-filtered through a 25 mm diameter, 0.3 nominal pore diameter glass fiber filter using a syringe and a plastic filter holder and stored in a locking petri dish in the dark and on ice until the filter was returned to the laboratory (within 6 hours). Subsequently, the sample was dried at 35-50 °C for a minimum of 24 hours. Following drying, the filters were stored in locking petri dishes in a desiccator.

Using the samples collected as described above, bacteria attached to suspended solids and those freely-suspended in water samples were differentiated by filtering samples through 2.0-µm pore size polycarbonate membrane filters, then cells in the filtrate (freely-suspended) and in unfiltered water were counted. Numbers of attached bacteria were determined by subtracting numbers of freely-suspended bacteria from the total bacteria present in unfiltered water.

The eelgrass bed located between Weeks Point and the channels at the middle of the Bay was the focus of preliminary studies for determining deposition/removal of suspended solids and/or microbial contaminants from the overlying waters of eelgrass beds. Aerial surveys were undertaken during July, 1991 and July, 1992 to map the distribution and extent of the bed. The characterization of eelgrass beds within the area of study included the assessment of eelgrass biomass, density, and reproductive tissues.

The shellfish beds on the southeastern shoreline of Adams Point were the focus of studies on the potential for shellfish to remove suspended bacteria. Size and dimensions of the oyster study area were established from a boat at low tide.

The current flow characteristics at specific tidal stages for both sites were established. Directional flow determinations were made to establish sites for water sampling. Sites were established for the eelgrass bed on a directional gradient for an incoming tide just after slack low tide with a first sample site in the channel in front of the bed, a second site at the edge of the bed, a third within the bed, and a fourth nearer to shore and further into the bed. Sites were established for the oyster bed on a directional gradient for an outgoing tide near slack low tide with a first sample site upstream in the mouth of Crommet Creek, a second over the bed, and a third downstream near the tip of Adams Point. At each study site, samples were analyzed for total suspended solids, % organic content, and bacterial indicators. Studies were repeated seasonally to demonstrate the varying effectiveness of the habitats caused by seasonally-related variations in growth/senescence of eelgrass and active filtration processes of oysters. The eelgrass study, including mesocosm work, was initiated in May, 1992, and has been continued as part of the continuation study during FY 92.

Flowing seawater tanks with added shellfish were used for artificially contaminating shellfish for determining the fate of microbial contaminants when flowing over a shellfish bed. Shellfish and water were analyzed for microorganisms at regular intervals following initial contamination.

RESULTS AND DISCUSSION

The routine surveys of water quality showed microbial contaminant concentrations to be temporally and spatially variable, as illustrated for enterococci (Figures 2-7). During the initial period of the project in the summer of 1991, indicator concentrations at AP and MB were very low, often < 10, and sometimes < 1, enterococci, FC, and Ec per 100 ml. At SR, levels were higher than at AP and MB, but they were still lower than observed for other times of year. Levels at all 3 sites increased in October and remained high, relative to summer samples, through early January. Indicator levels were generally lower again in late spring/early summer of 1992, especially at AP and MB. The seasonal trends observed are typical for Great Bay, and probably reflect a variety of influencing factors. Concentrations are probably lowest in summer because of higher temperatures, as fecal bacteria survive best at lower temperatures (24). The activities of zooplankton and the more competitive indigenous bacteria are also greater in the warmer waters during summer. Protistan grazing of bacteria is a major factor controlling levels of indicators in natural waters (25). Other hydrological, meteorological, physical and biological factors may also be important influencing factors.

As previously discussed, levels of microbial contaminants are typically higher in the Squamscott River than in the Great Bay, despite recent upgrading of POTW facilities that discharge into the river. The results of the routine monitoring of levels at the three stations showed that the levels of microorganisms at SR were higher than levels in the Bay at MB and AP (Figures 8-15). Comparisons of averages for all data collected for each site showed SR to always have significantly higher levels of all four indicators at both low and high tides (Table 1). Levels of indicators were not different at MB and AP at low tide. However, levels of FC, Ec and enterococci were significantly lower at MB than at AP at high tide. This is consistent with the idea that greater levels of contaminants enter Great Bay with tidal waters from the more contaminated waters downstream, and natural processes may decrease levels as the water flows through Great

Bay past AP. Levels of the ever-present and conservative fecal tracer, Cp, were not different at AP and MB at high tide.

In general, the concentrations of TSS were highest in the Squamscott River and decreased rapidly in a bayward direction (Figure 16). TSS concentrations ranged from 2.5 to 37.6 mg/l at AP, with the highest concentrations occurring in spring, 1992, and the lowest in fall, 1991. The tidal currents at Adams Point are very strong and the water column is normally relatively vertically mixed. Consequently, there is not a strong vertical gradient for TSS concentrations. This is, in part, due to sampling close to low or high tide periods which reduces the impact of bottom sediment resuspension. In addition, during periods when we did not sample at slack water, the strong tidal currents caused the submersible pump to swing out, causing the sample to be collected further from the bottom. Consequently, we did not always obtain a true near-bottom sample.

TSS concentrations at MB varied from 2.2 to 38.2 mg/l, with highest values again occurring in spring, 1992. This maximum concentration occurred close to the bottom and is attributed to resuspension. However, TSS concentrations generally did not consistently increase with depth as expected. The water depths are less and the current velocities generally lower at the mid-bay station than at AP. It is not clear at this point why the concentrations did not increase with depth, unless resuspension is less important than expected at this site or the low currents associated with low or high tide periods allowed resuspended particles to settle out.

At SR, TSS concentrations ranged from 6.4 to 64.5 mg/l, with highest concentrations again occurring during spring, 1992. However, sampling was not done directly following Hurricane Bob in this data set, which had the highest TSS concentrations (see the surface water samples discussed below). This station is located in an estuarine tributary which introduces high riverine inputs to the system. Similar to the station at AP, the currents are strong at SR, causing the water column to be vertically mixed.

In addition to the submersible pump sampling, TSS concentrations were also determined on hand-collected surface samples. The distribution of the TSS load as indicated by these surface

samples showed the same patterns as just described (Figure 17). The TSS concentrations ranged from 2.4 to 61.9 mg/l at AP, from 2.1 to 20.3 at MB, and 2.5 to 175.1 mg/l at SR. Concentrations were highest at SR, and at all stations during low tide periods.

On two occasions, more intensive sampling of the water from MB to CL, via the channel of Great Bay that leads to and includes the mouth of the Squamscott River, was undertaken at low tide to measure the gradient of contaminants from CL. The results of the first sampling showed a sharp gradient of concentrations to occur close to the mouth of the Squamscott River. On the second occasion, contaminant levels were elevated all along the sampling transect, probably because of the presence of hundreds of migratory birds that were present and contributing fecal wastes on the surrounding mudflats at low tide. The details of the natural processes that are associated with the decrease in microbial contaminants at the mouth of the Squamscott River are the focus of a new project.

The stage of the tide also had an influence on microbial levels. Indicators levels at AP were typically higher at high tide than at low tide (Table 1), although this trend was not significant (Table 2). The trend at AP is consistent with previous findings over the previous three years, and supports the idea that water leaving Great Bay at AP typically has lower levels of microbial contaminants than water entering the Bay at high tide. The opposite trend was observed at SR and MB. Levels of all indicators were higher at low tide at SR; these differences were highly significant for enterococci, FC and Ec (Table 2). This trend is expected, as water at low tide should reflect the greatest influence of contaminated freshwater sources of pollution. The only significant deviations from the concentrations of fecal-borne microorganisms being higher at low tide occurred during the days following Hurricane Bob, in which case levels were much higher than normal at both tides. At MB, levels of all indicators were also higher at low tide; the differences for FC and Ec were significant (Table 2). The reasons for this trend at MB may reflect the integrated influences of contaminant removal processes as water flows over the extensive eelgrass and oyster beds located throughout the middle shallow portions and the channels of the

Bay. The higher levels at low tide probably reflect the greater levels of contaminants associated with the more polluted freshwater from the Lamprey and Squamscott rivers that flow into the Bay and have a greater influence at low tide.

Hydrological and meteorological events had significant influences on concentrations of microbial contaminants in Great Bay. The most striking influence on microbial concentrations came in response to the heavy rains caused by Hurricane Bob in August, 1991. Hurricane Bob caused over 6 inches of rainfall in the watershed on August 19-20, increasing microbial and TSS concentrations dramatically and decreasing salinities. For example, enterococci levels increased at all sites from background levels, which were ~1/100 ml on August 13, to >500 on August 21 (Figure 18). Levels remained high (enterococci >25/100 ml) for at least 4 days following the first day of the storm as contamination continued to flow into the Bay and contaminant levels within the Bay had not yet attenuated. Contaminant levels returned to the characteristic low levels after 7-14 days. Less extreme increases in levels were also observed in November following two separate heavy rain storms (Figures 2-7).

Maximum TSS concentrations (175.1 mg/l) occurred in the Squamscott River on August 20 directly following the hurricane (Figure 19). TSS concentrations decreased rapidly, reaching 19.1 mg/l at AP on this same day. TSS concentrations at AP directly following the storm were higher than normal, however, far below those measured at SR. No MB sample was collected on August 20 due to problems associated with the hurricane. Also, the samples taken at AP and at SR were taken from close to shore which may have accounted for some of the higher TSS concentrations. The high TSS concentrations associated with the storm decreased rapidly within a few days either by deposition or dilution.

A substantial database has been built up on synoptic levels of microbial contaminants and total suspended solids for characterizing the relationship between the two measurements. In general, both total suspended solid and microbial contaminant concentrations were routinely higher at CL than at MB or FS. However, there are no general trends between suspended solids and

indicator concentrations (Figures 20-29). There are apparent differences between different indicators and between sites. No relationships between indicators and suspended solids were apparent for any indicator at AP and MB (Figures 20-23). There were better apparent relationships at SR, especially at HT, for enterococci, FC and Cp (Figures 24-29), with greater levels of indicators being present with higher concentrations of suspended solids. A number of factors could account for these observations, including salinity, tidal stage, organic content of the suspended sediments, and the proportion of adsorbed bacteria, as discussed below. A larger database is needed, and is presently being generated, to allow for statistically-sound analyses of these data.

Studies on numbers of freely-suspended and particle-associated indicators showed some somewhat unexpected results. Much of the time, the analyses indicated that almost all of the fecal coliforms and, especially, enterococci were not associated with particles. However, some of the data also suggest that the filtration process may have some influence on packets of enterococci, because numbers of free-living enterococci (those that pass a 2.0 μm filter) were often higher than total enterococci, the data indicating 120% of enterococci cells were freely suspended (Table 3). The fecal coliforms were apparently more strongly or consistently bound to particles, as only 41% of the cells were freely suspended. The limited data did suggest that in highly contaminated waters, a much greater proportion of fecal coliforms were freely suspended. The implications of these results and alternative methods for enumeration of particle-bound enterococci and fecal coliforms in estuarine water are presently under investigation. A different indicator, C. perfringens, was used starting in the spring of 1992 at the three stations. Results indicate a strong association of C. perfringens spores with suspended particles $>2.0 \mu\text{m}$. Continued analysis showed this relationship to be consistent at all concentrations, and overall $>95\%$ of C. perfringens cells were associated with suspended particles. The different responses between indicators may help to explain some of the data on the relationship between suspended sediments and indicator levels illustrated in Figures 16-25.

The differences between indicators is probably a function of methodology, environmental factors, and physiological differences between indicators. Probably all of the C. perfringens cells detected were in fact spores in the estuary. Enterococci are small gram positive cocci that probably exist as packets of cells, as previously discussed. The fecal coliforms and E. coli are gram negative bacteria that are probably present as relatively dormant small cocci (Roszak and Colwell, 1987). Thus, the success of using C. perfringens was probably a function of these factors not being problems. The strong relationship between C. perfringens and particles was exploited in ensuing studies on the effects of eelgrass beds on water column concentrations of microbial contaminants.

Other research focused on the natural mechanisms that could be causing contaminant levels to be lower as water flows across Great Bay. Research focused on shellfish and eelgrass beds at various locations in Great Bay. The dimensions of the oyster bed located on the southeastern shoreline of Adams Point was determined. Studies of microbial contaminant levels upstream, over, and downstream of this shellfish bed did not show any consistent evidence of removal of microorganisms. Studies in mesocosm tanks showed oysters reduced levels of suspended bacteria from water by a factor of at least 10^3 within 15 minutes, and maintained low levels thereafter. The reasons for little detectable effect of oysters on microbial concentrations in the field were probably a result of sampling water too far above the oysters, levels of contaminants in the water too low to show large enough differences, and the heavier than usual recreational harvest of oyster from the bed during the fall of 1991 and spring of 1992. The latter factor was exacerbated by the recent closing of the softshell clam beds in the coastal New Hampshire estuaries, leaving Great Bay as the only remaining area in New Hampshire where shellfish harvesting is legal.

Aerial surveys were undertaken during the peak growing season to map the distribution and extent of the eelgrass beds in the Bay. The eelgrass beds located to the west of the main channel of Great Bay just south of AP were the focus of initial studies for determining deposition/removal of suspended solids and/or microbial contaminants from the overlying waters of eelgrass beds.

Current flow was measured during a falling tide and water samples taken parallel to current flow upstream, over, and downstream of the bed. Initial results, based on enterococci levels and conducted during the fall of 1991 at times when levels were relatively low, showed a small removal effect on enterococci in water flowing through the bed.

More intensive studies were conducted during the spring of 1992 on a large eelgrass bed located in the south central portion of the Bay, just south of the fork in the main channel, between Weeks Point and the channels at the middle of the Bay. The bed was characterized by assessing biomass, density, and reproductive tissues (Table 4). Samples were collected from overlying waters along a transect that paralleled water flow direction through the bed. Sites were established for the eelgrass bed on a directional gradient for an incoming tide just after slack low tide, with a first site in the channel just in front of the bed, a second site at just within the edge of the bed, and third and fourth sites approximately 100 and 200 meters further into the bed. The study was designed to document potential removal of bacteria by settlement within the eelgrass bed. Thus, replicate water samples were collected at the four sites along the transect on calm days just after slack tide to avoid the potentially elevated contaminant levels associated with resuspension of sediments by wave action.

Analyses, mainly of C. perfringens (Table 5), showed eelgrass beds may be effective in reducing contaminant levels. The averages for five sampling dates were 13, 11, and 7 C. perfringens /100 ml for the channel, bed edge, and within-bed sites. Most of the C. perfringens removed were probably associated with suspended particles, as numbers of freely-suspended cells were similar for water samples from within and outside of the bed. The ongoing project has continued this study under a wider set of conditions.

SUMMARY

This study has confirmed previously-observed trends that indicate that microbial

contaminant levels decrease as water flows through Great Bay. Levels of contaminants in water from the tributaries and from downstream areas were higher than levels in the middle of the Bay. Suspended sediment concentrations also decreased rapidly in a bayward direction from the Squamscott River to the middle of the Bay. Concentrations of both microbial contaminants and TSS increased rapidly following the intense rains associated with Hurricane Bob. The recovery of the estuary was relatively rapid for TSS, but slower for microbial contaminants at all three sites. This and other observations illustrate the apparently complex relationship between suspended sediments and the target bacteria.

Little direct relationship was apparent between indicator and TSS concentrations for most conditions studied. Some of this was probably a function of the different levels of association for the different indicators and TSS, which may be related to the physiological and morphological differences between the indicators. Whereas we found no evidence of enterococci being attached to particles using our analytical procedure, C. perfringens cells appeared to be almost entirely associated with particles. A larger database will be useful to determine what factors may be causing differences between indicators and what properties of suspended sediments and estuarine environments may govern interactions between bacteria and suspended particles.

The most useful information from this study may be the preliminary indications that microbial contaminants in water flowing over eelgrass beds may indeed settle out of the water column under calm conditions. Continuing studies will help to confirm these observations to give a better assessment of the importance of this and other natural processes in reducing public health risks associated with the fecal contamination of estuarine water.

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Table 1. Ranked comparison of geometric means of indicator levels (cfu/100 ml) at 3 sites using Duncan new multiple range test. Significance level = 0.05.

| Site | Enterococci | | Fecal coliforms | | <i>Escherichia coli</i> | | <i>Clostridium perfringens</i> | |
|------------------|-------------|-----------|-----------------|-----------|-------------------------|-----------|--------------------------------|-----------|
| | High tide | Low tide | High tide | Low tide | High tide | Low tide | High tide | Low tide |
| | Mean/rank | Mean/rank | Mean/rank | Mean/rank | Mean/rank | Mean/rank | Mean/rank | Mean/rank |
| Adams Point | 3.7 b | 2.5 a | 16 b | 8.0 a | 10 b | 6.6 a | 8.7 a | 8.9 a |
| Mid Bay | 1.4 a | 1.7 a | 3.9 a | 12.0 a | 3.2 a | 9.8 a | 7.8 a | 10.0 a |
| Squamscott River | 9.5 c | 49 b | 23 b | 159 b | 16 b | 111 b | 20 b | 34 b |

Table 2. Significance levels for comparison of indicator levels at high and low tides using paired t tests.

| Indicator | Adams Point | Sites | |
|--------------------------------|-------------|---------|---------------|
| | | Mid Bay | Squamscott R. |
| Enterococci | 0.24 | 0.29 | 0.0002** |
| <i>Clostridium perfringens</i> | 0.44 | 0.27 | 0.2 |
| <i>Escherichia coli</i> | 0.91 | 0.05* | 0.002** |
| Fecal coliforms | 0.32 | 0.05* | 0.005** |

Table 3. Proportion of indicator bacterial cells not attached to particles (>2.0 μm diameter) in estuarine water.

| Indicator | Total cells | Freely-suspended cells | % cells freely-suspended | |
|-----------------------|-------------|------------------------|--------------------------|-------|
| <i>C. perfringens</i> | 332 | 16 | 4.8 | n= 17 |
| Fecal coliforms | 46 | 19 | 41 | n= 7 |
| Enterococci | 159 | 193 | 121 | n= 17 |

Table 4. Eelgrass standing stock data for Weeks Point (WP) bed in Great Bay: 1991-92.

| Station | Date | Density #/m ² ± se | Reproductive #/m ² ± se | Spathes #/m ² ± se | Leaf biomass g/m ² ± se | Flower biomass g/m ² ± se | Root biomass g/m ² ± se |
|---------|--------|----------------------------------|---------------------------------------|----------------------------------|---------------------------------------|---|---------------------------------------|
| WP | 7/8/91 | 174 ± 18 | 0 | 0 | 76 ± 11 | 0 | 0 |
| WP | 7/8/91 | 254 ± 34 | 22 ± 9.5 | 1.2 ± 0.4 | 272 ± 39 | 101 ± 35 | 0 |
| WP-A | 7/6/92 | 309 ± 19 | 2.7 ± 2.7 | 0 | 242 ± 11 | 13 ± 0 | 32 ± 2.7 |
| WP-B | 7/6/92 | 376 ± 55 | 0 | 0 | 352 ± 30 | 0 | 45 ± 8.1 |
| WP-C | 7/7/92 | 325 ± 51 | 0 | 13 ± 13 | 245 ± 28 | 4.8 ± 0 | 33 ± 5.0 |

Table 5. *Clostridium perfringens* concentrations at sites in and around an eelgrass bed in Great Bay: May-June, 1992.

| | Channel | Site location | |
|--------------------|---------|---------------|------------|
| | | Bed edge | Inside bed |
| Geometric mean | 13.3 | 10.9 | 6.9 |
| Standard deviation | 6.9 | 4.8 | 5.8 |
| n= | 5 | 5 | 8 |

FIGURE LEGENDS

Figure 1. Map of the Great Bay Estuary showing the three sampling sites (X) at Adams Point (AP), Mid Bay (MB) and the Squamscott River (SR).

Figure 2. Enterococci levels at Adams Point at high tide-6/91 to 8/92.

Figure 3. Enterococci levels at Adams Point at low tide-6/91 to 8/92.

Figure 4. Enterococci levels at Mid Bay at high tide-6/91 to 8/92.

Figure 5. Enterococci levels at Mid Bay at low tide-6/91 to 8/92.

Figure 6. Enterococci levels in the Squamscott River at high tide-6/91 to 8/92.

Figure 7. Enterococci levels in the Squamscott River at low tide-6/91 to 8/92.

Figure 8. Geometric mean enterococci concentrations at high tide-6/91 to 8/92.

Figure 9. Geometric mean enterococci concentrations at low tide-6/91 to 8/92.

Figure 10. Geometric mean fecal coliform concentrations at high tide-6/91 to 8/92.

Figure 11. Geometric mean fecal coliform concentrations at low tide-6/91 to 8/92.

Figure 12. Geometric mean E. coli concentrations at high tide-6/91 to 8/92.

Figure 13. Geometric mean E. coli concentrations at low tide-6/91 to 8/92.

Figure 14. Geometric mean C. perfringens concentrations at high tide-6/91 to 8/92.

Figure 15. Geometric mean C. perfringens concentrations at low tide-6/91 to 8/92.

Figure 16. TSS concentrations in Great Bay: pump samples-low tide-surface.

Figure 17. TSS concentrations in Great Bay: hand samples-low tide-surface.

Figure 18. Enterococci concentrations at 3 sites in Great Bay before, during, and after Hurricane Bob: August 19-20, 1991.

Figure 19. TSS-Hurricane Bob: hand samples-low tide-surface.

Figure 20. Relationship between enterococci and suspended sediments at Adams Point at high tide.

Figure 21. Relationship between enterococci and suspended sediments at Adams Point at low tide.

Figure 22. Relationship between enterococci and suspended sediments in Mid Bay at high tide.

Figure 23. Relationship between enterococci and suspended sediments in Mid Bay at low tide.

Figure 24. Relationship between enterococci and suspended sediments in the Squamscott River at high tide.

Figure 25. Relationship between enterococci and suspended sediments in the Squamscott River at low tide.

Figure 26. Relationship between fecal coliforms and suspended sediments in the Squamscott River at high tide.

Figure 27. Relationship between fecal coliforms and suspended sediments in the Squamscott River at low tide.

Figure 28. Relationship between *C. perfringens* and suspended sediments in the Squamscott River at high tide.

Figure 29. Relationship between *C. perfringens* and suspended sediments in the Squamscott River at low tide.

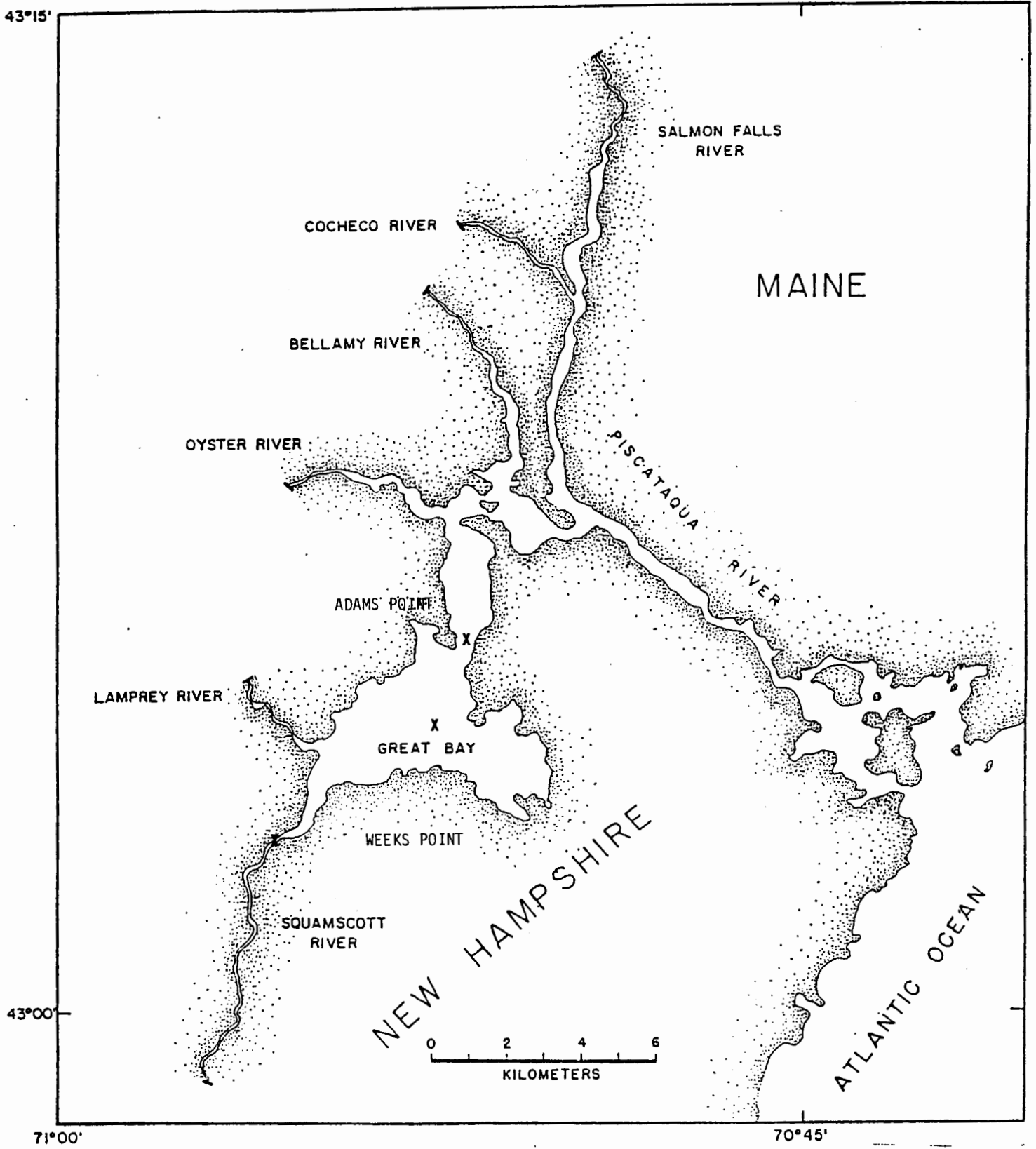


Figure 1. Map of the Great Bay Estuary showing the three sampling sites (X) at Adams Point (AP), Mid Bay (MB) and the Squamscott River (SR).

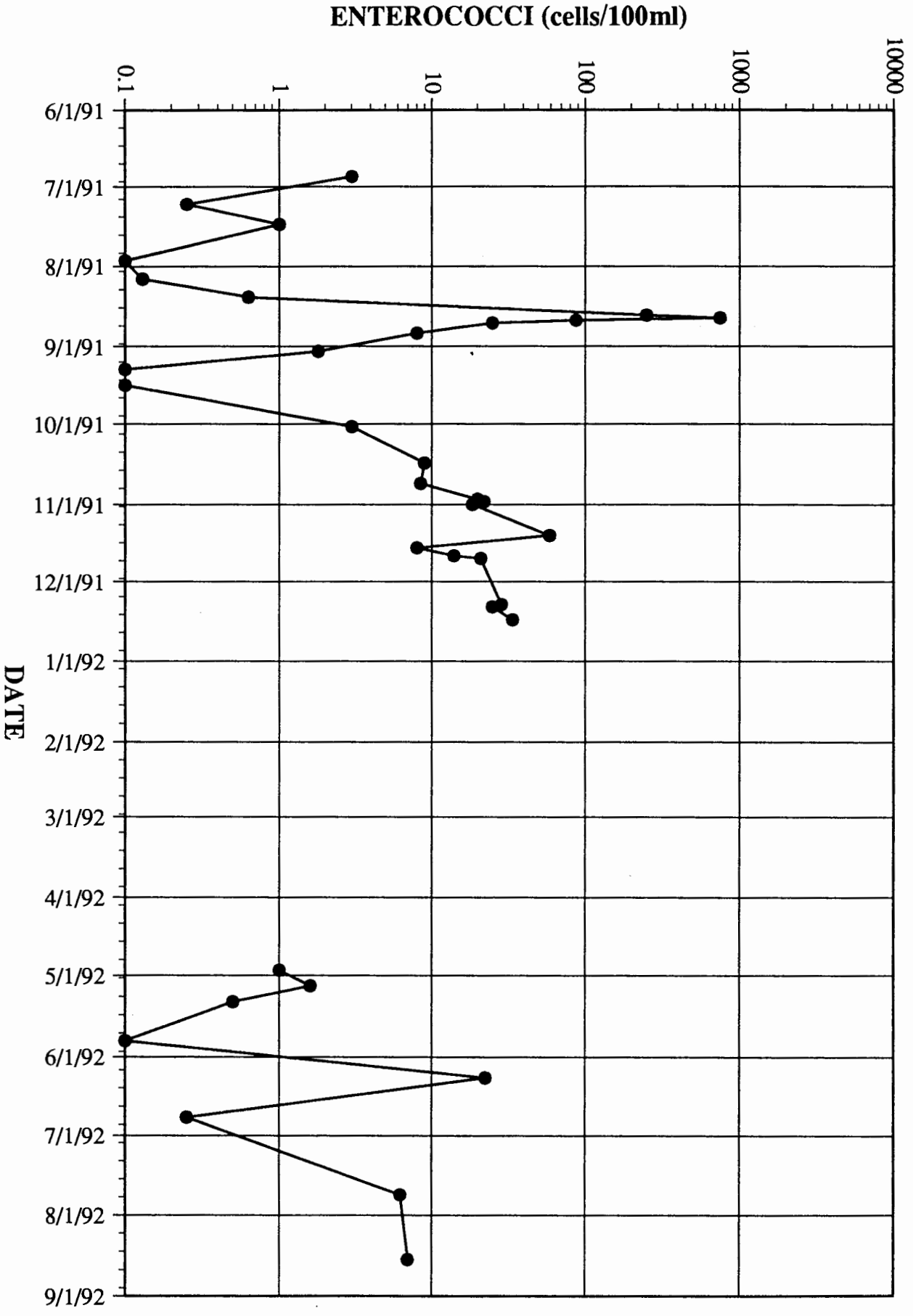


Figure 2. Enterococci levels at Adams Point at high tide - 6/91 to 8/92.

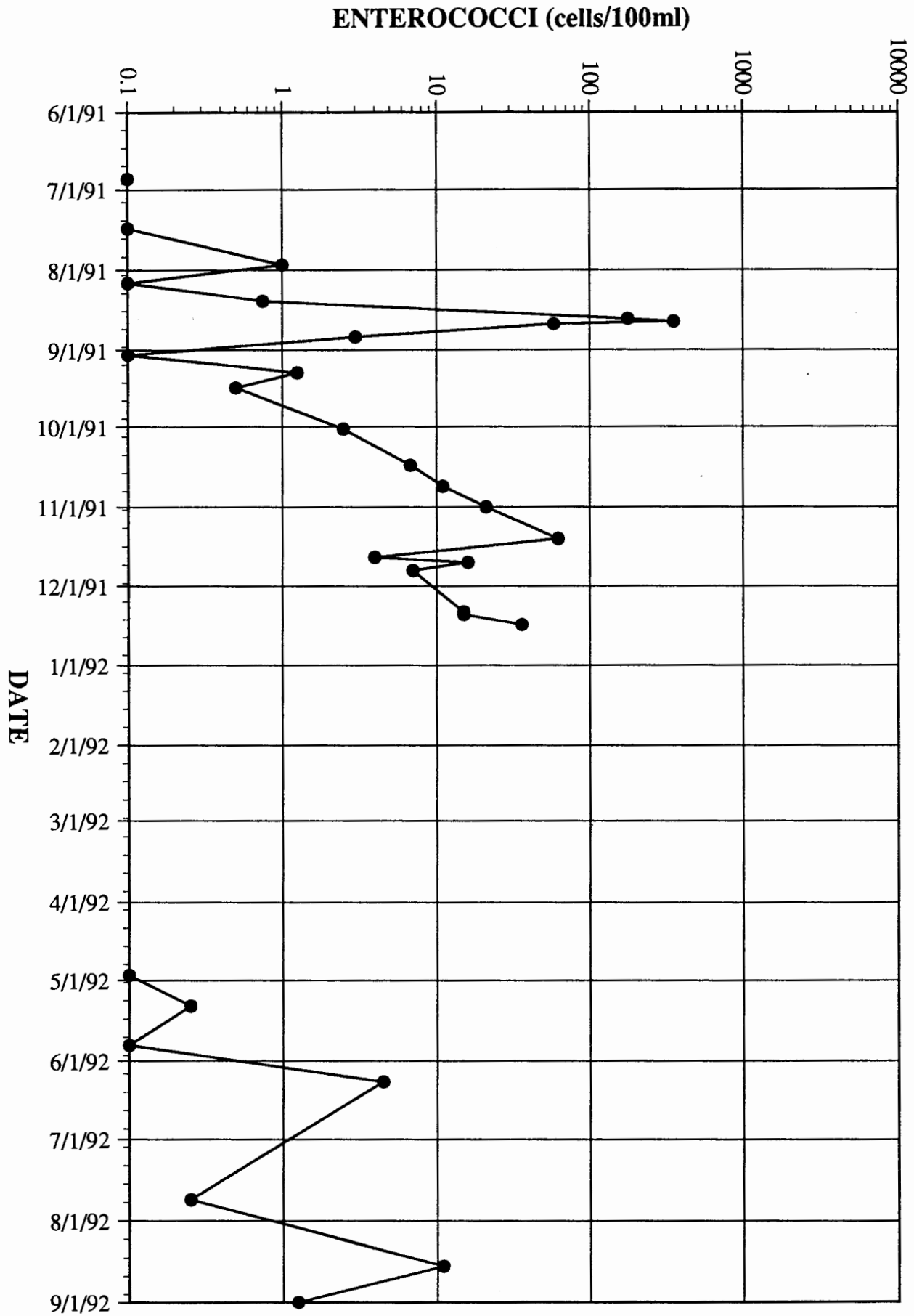


Figure 3. Enterococci levels at Adams Point at low tide - 6/91 to 8/92.

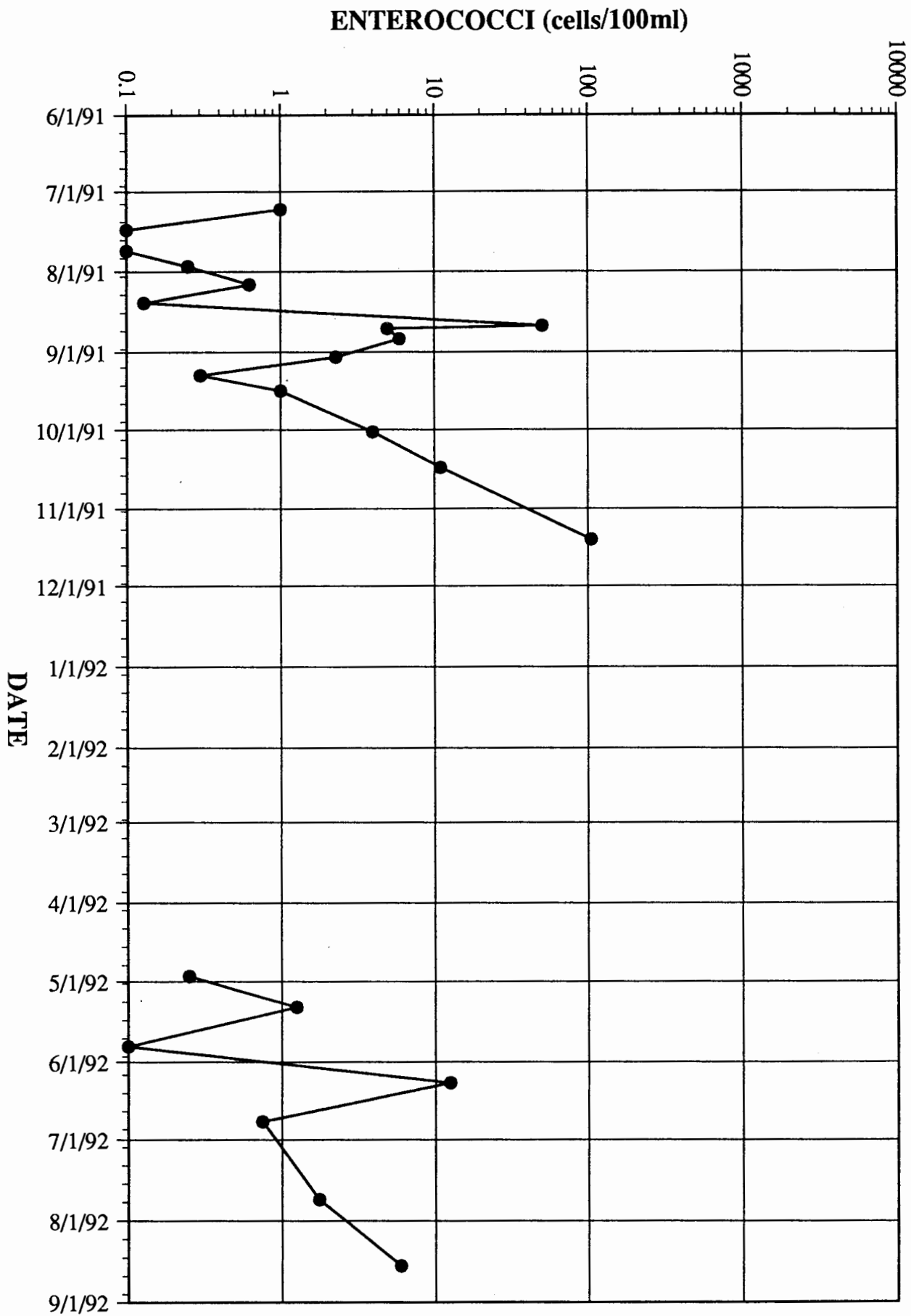


Figure 4. Enterococci levels in Mid Bay at high tide - 6/91 to 8/92.

ENTEROCOCCI (cells/100ml)

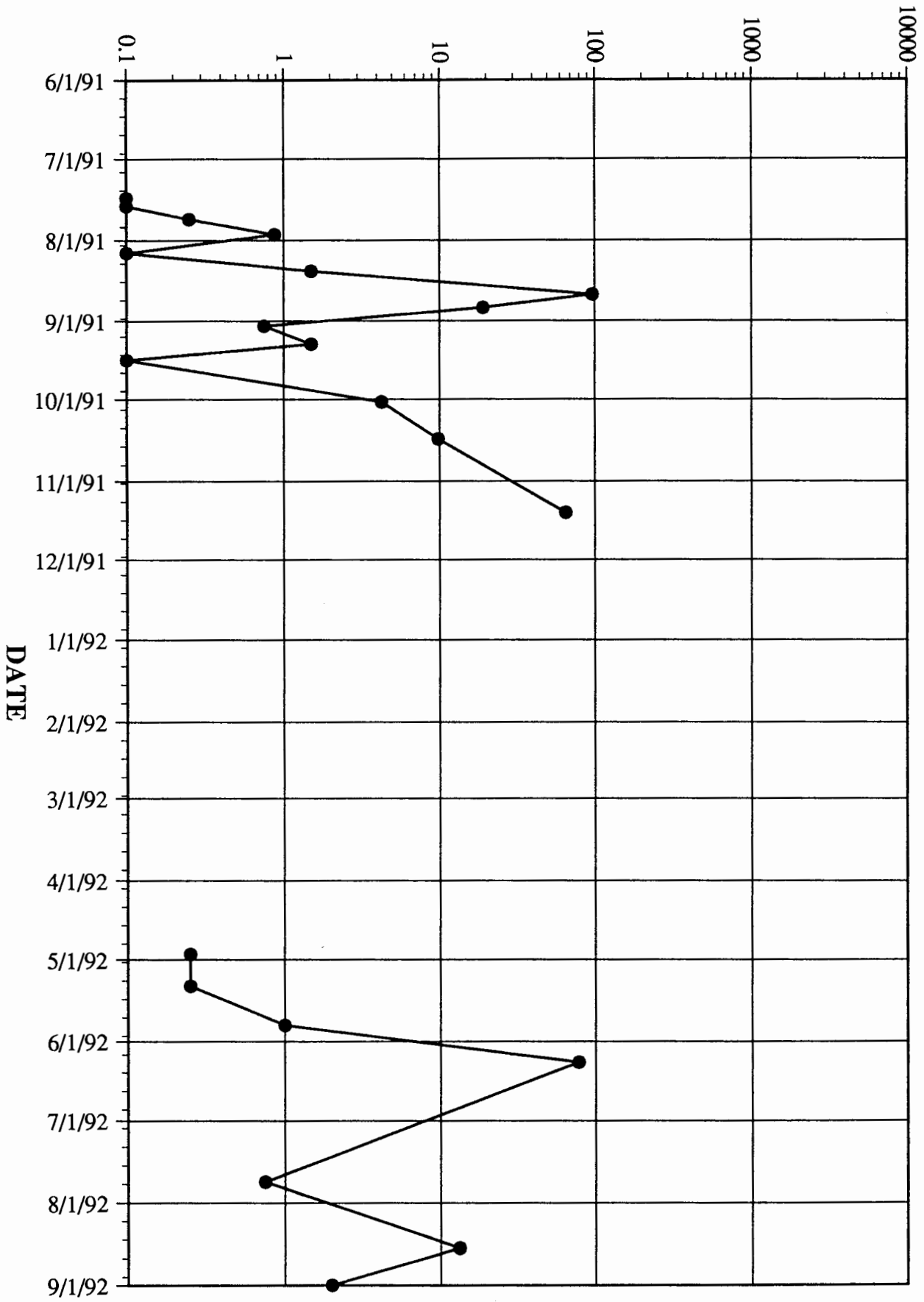


Figure 5. Enterococci levels in Mid Bay at low tide - 6/91 to 8/92.

ENTEROCOCCI (cells/100ml)

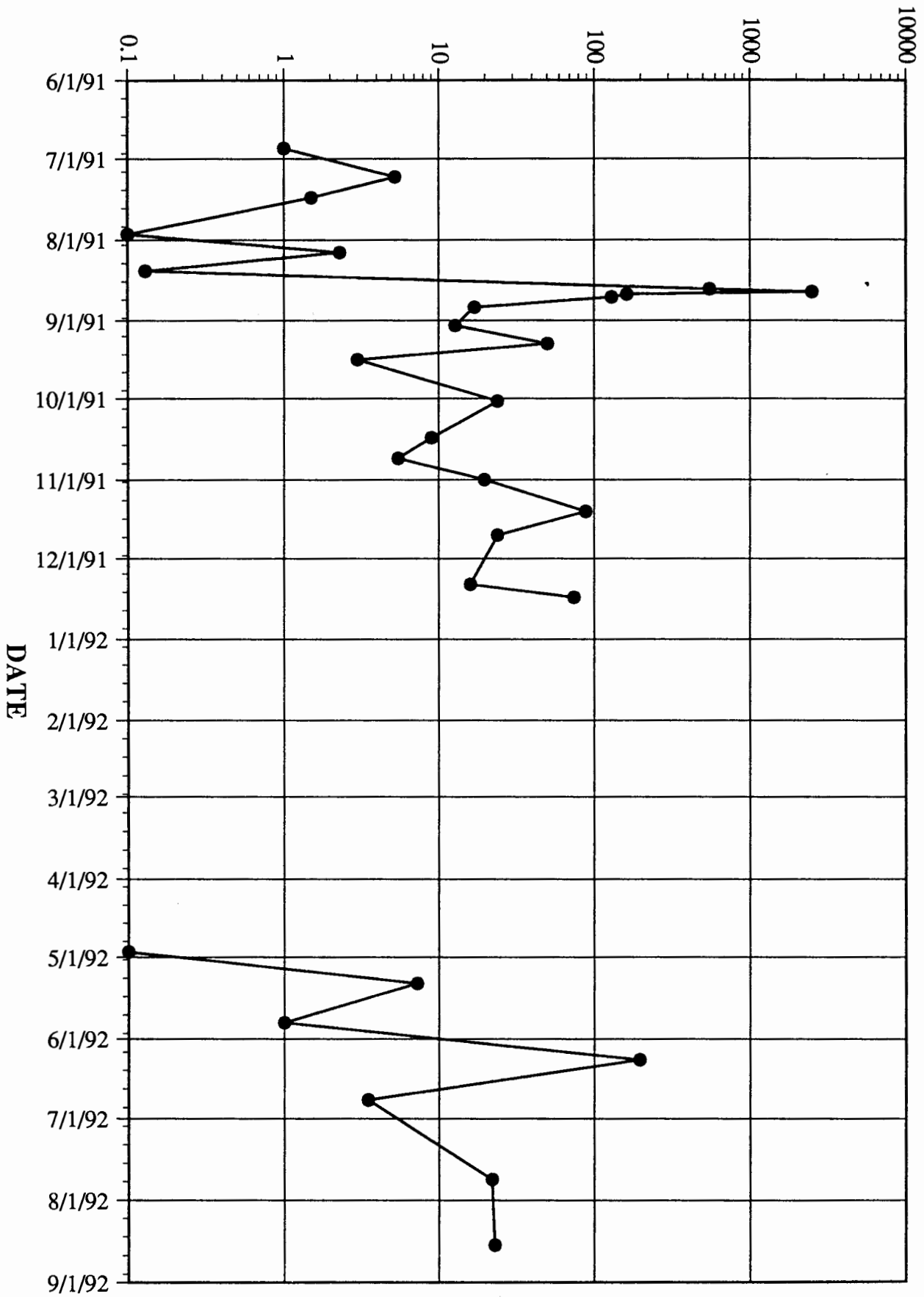


Figure 6. Enterococci levels in the Squamscott River at high tide - 6/91 to 8/92.

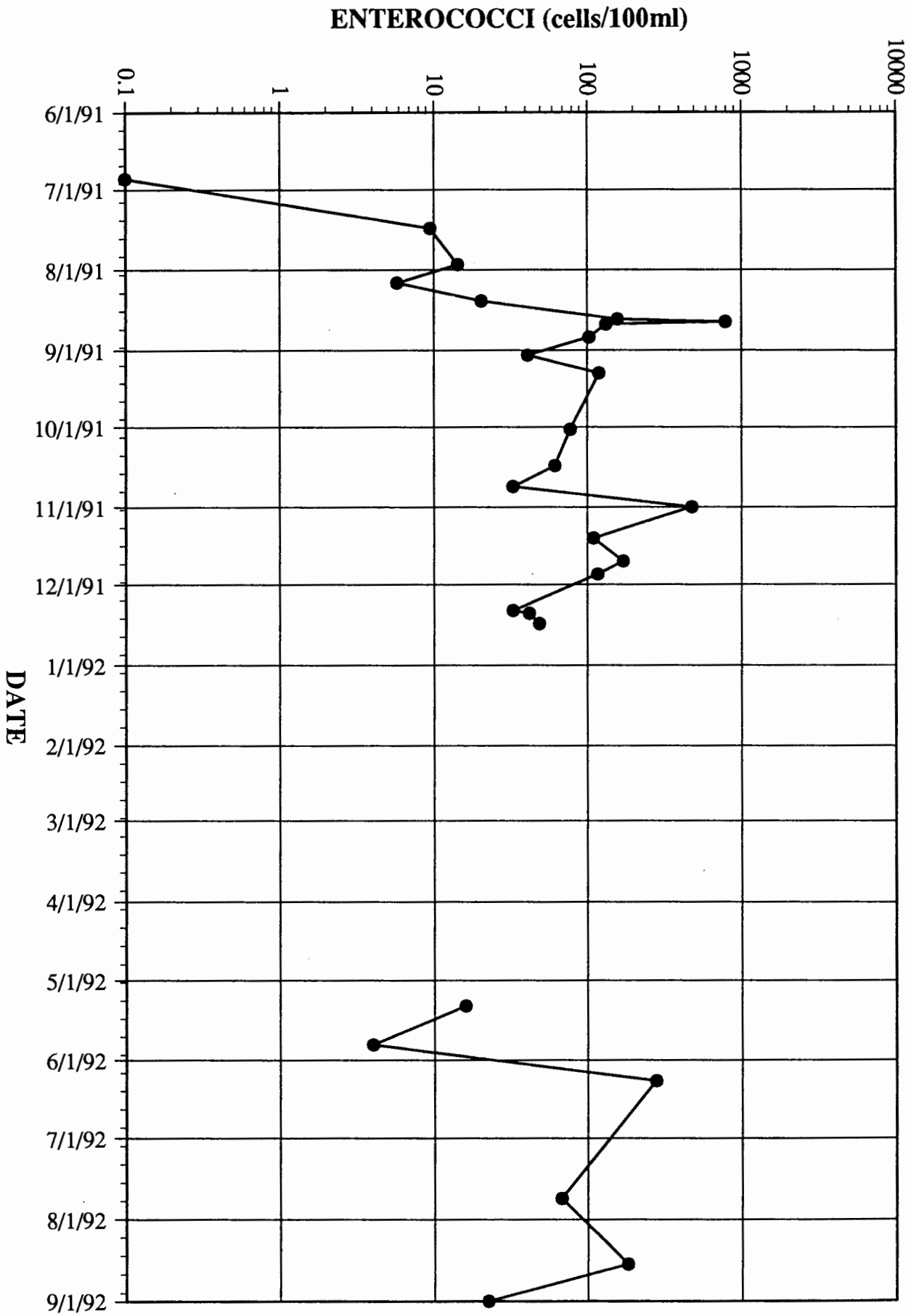


Figure 7. Enterococci levels in the Squamscott river at low tide - 6/91 to 8/92.

Figure 8. Geometric mean Enterococci concentrations at high tide
6/91 to 8/92.

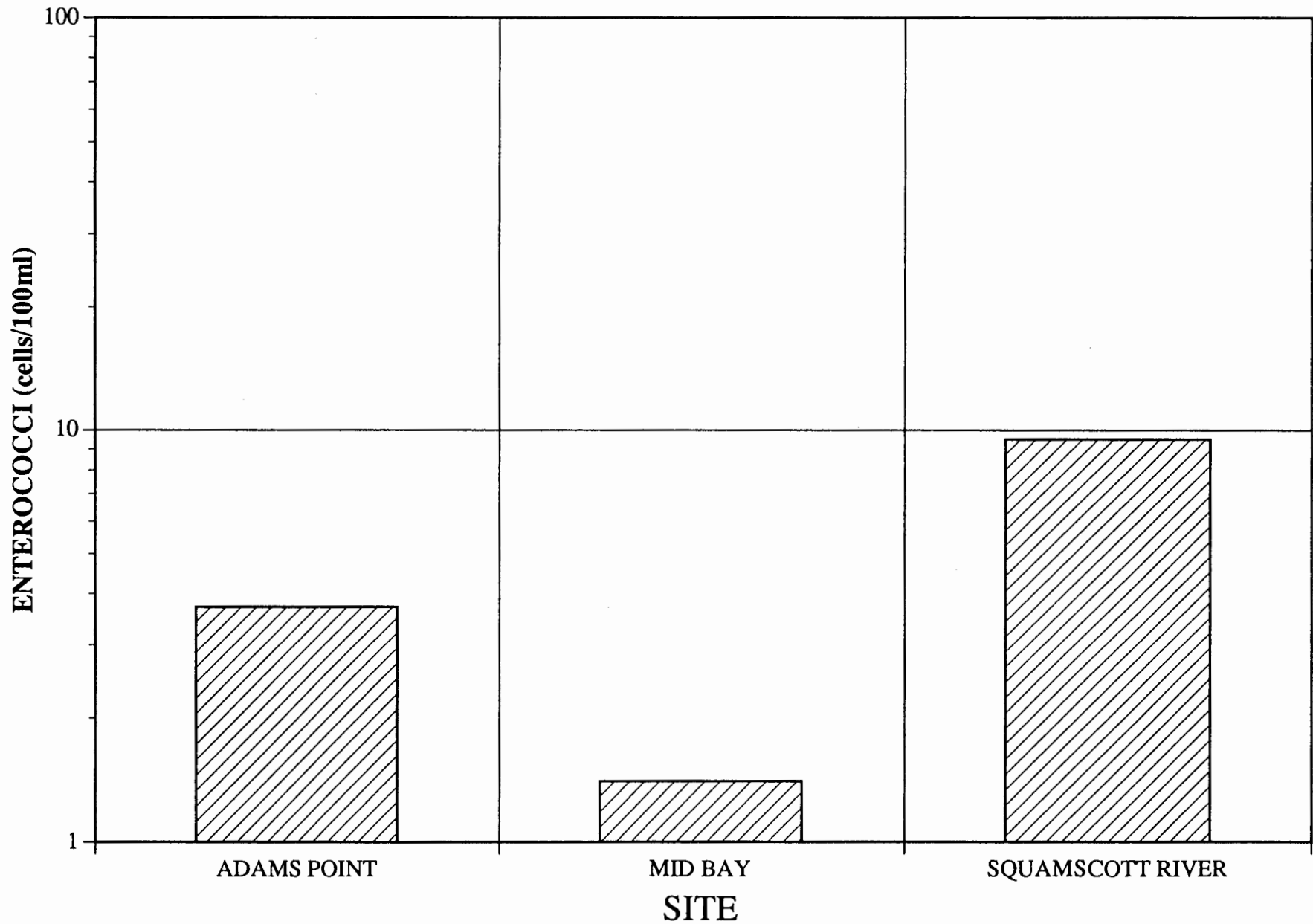


Figure 9. Geometric mean Enterococci concentrations at low tide
6/91 to 8/92.

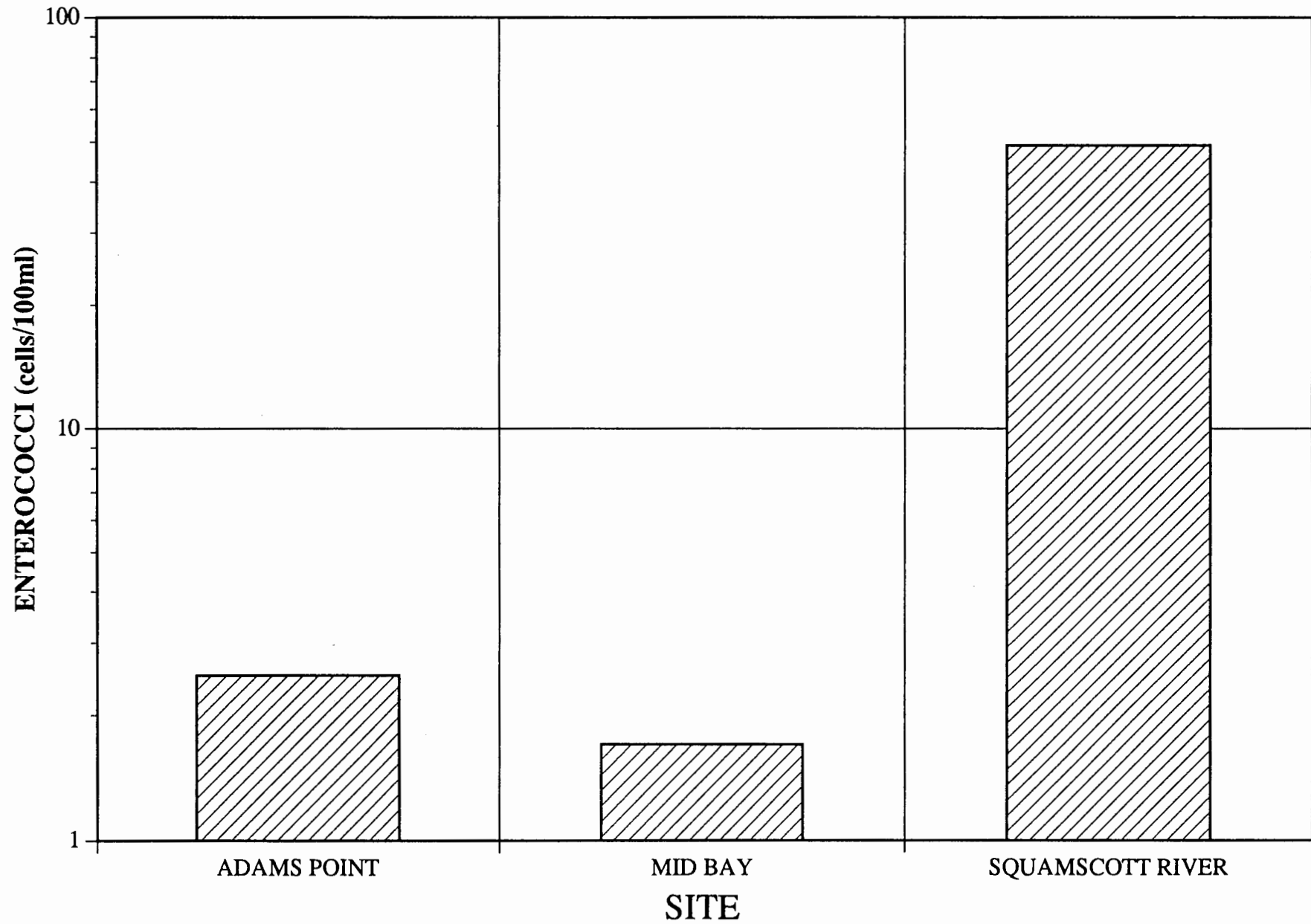


Figure 10. Geometric mean fecal coliforms concentrations at high tide
6/91 to 8/92.

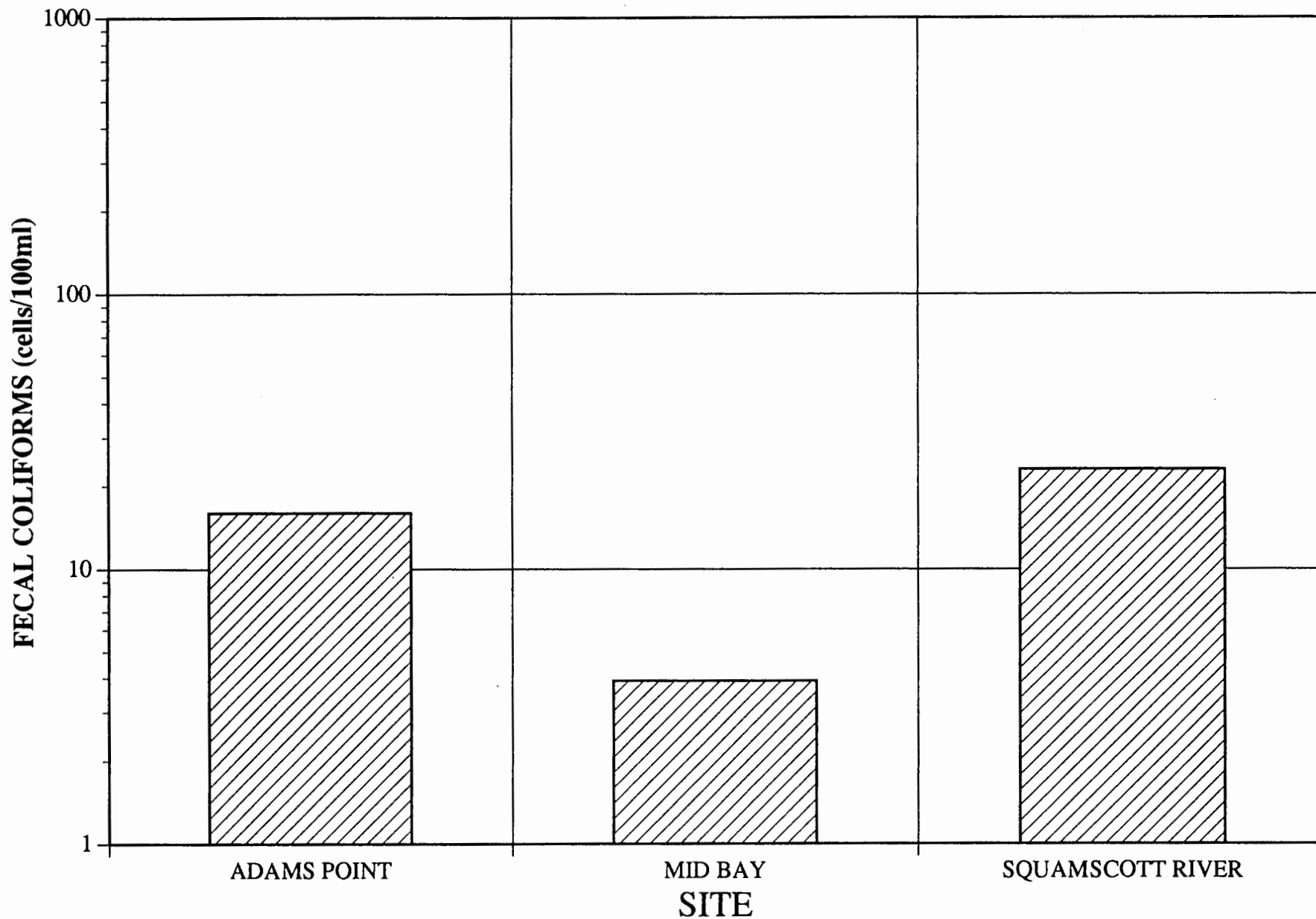


Figure 11. Geometric mean fecal coliforms concentrations at low tide
6/91 to 8/92.

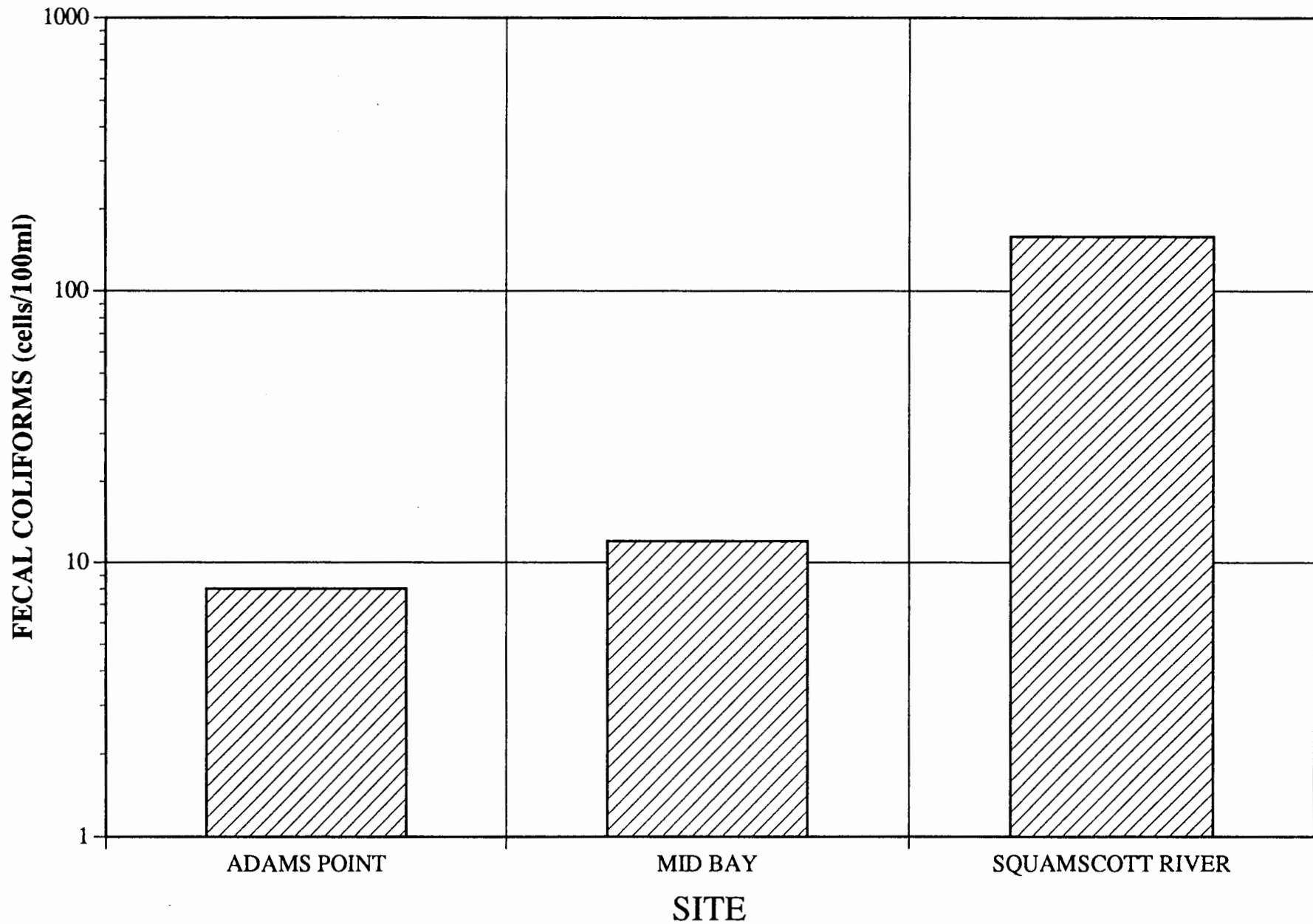


Figure 12. Geometric mean E. coli concentrations at high tide 6/91 to 8/92.

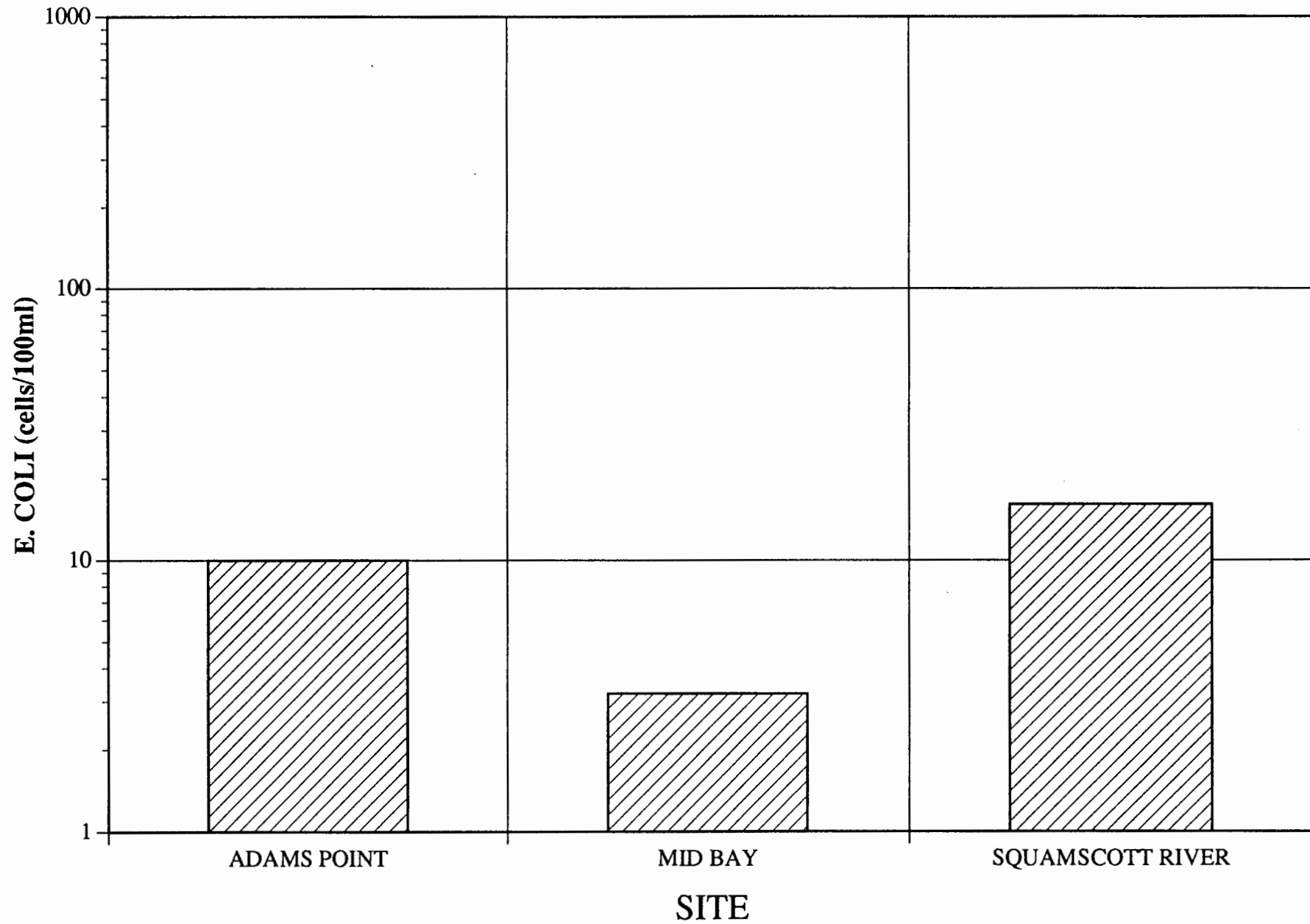


Figure 13. Geometric mean E. coli concentrations at low tide 6/91 to 8/92.

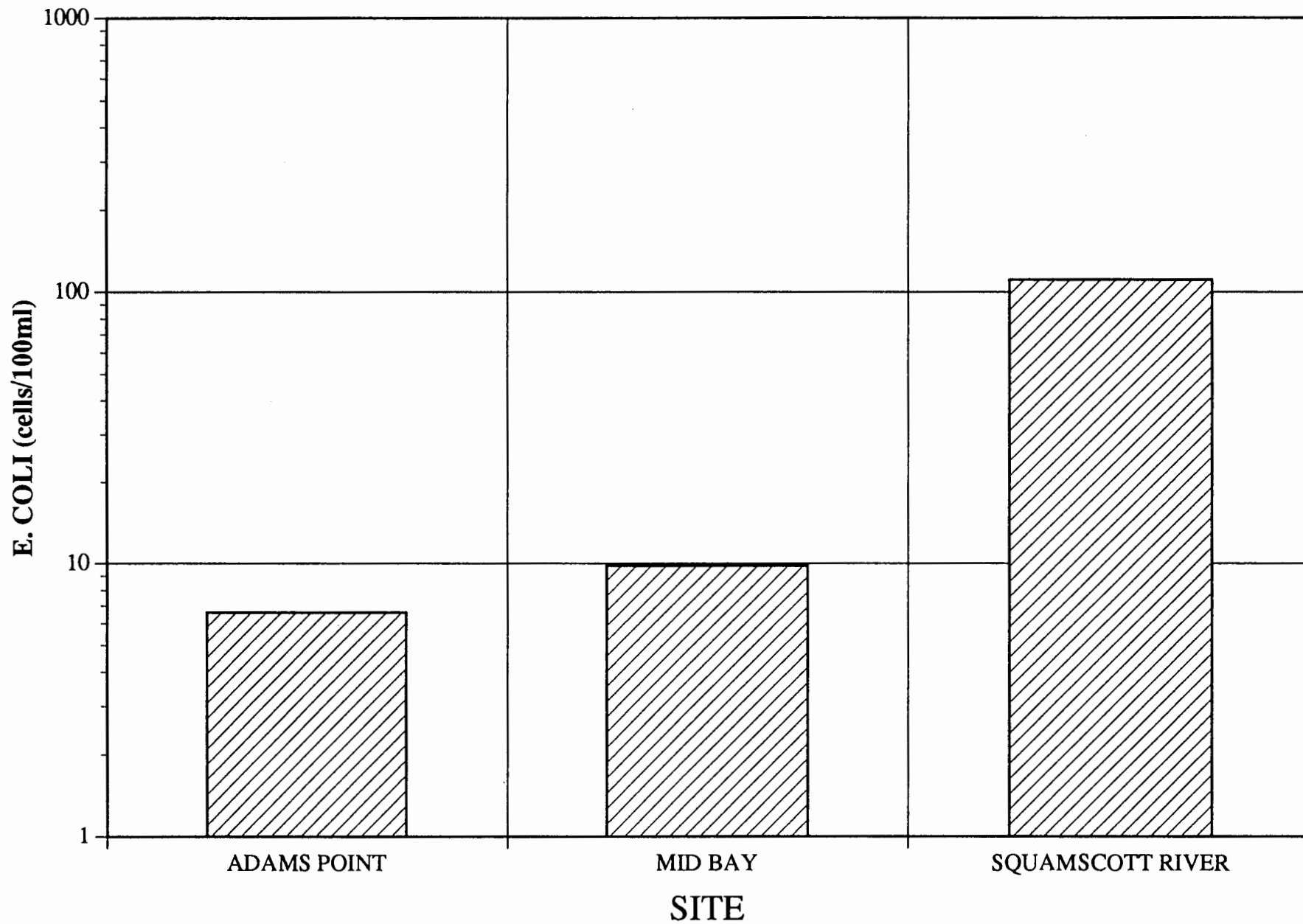


Figure 14. Geometric mean *C. perfringens* concentrations at high tide
6/91 to 8/92.

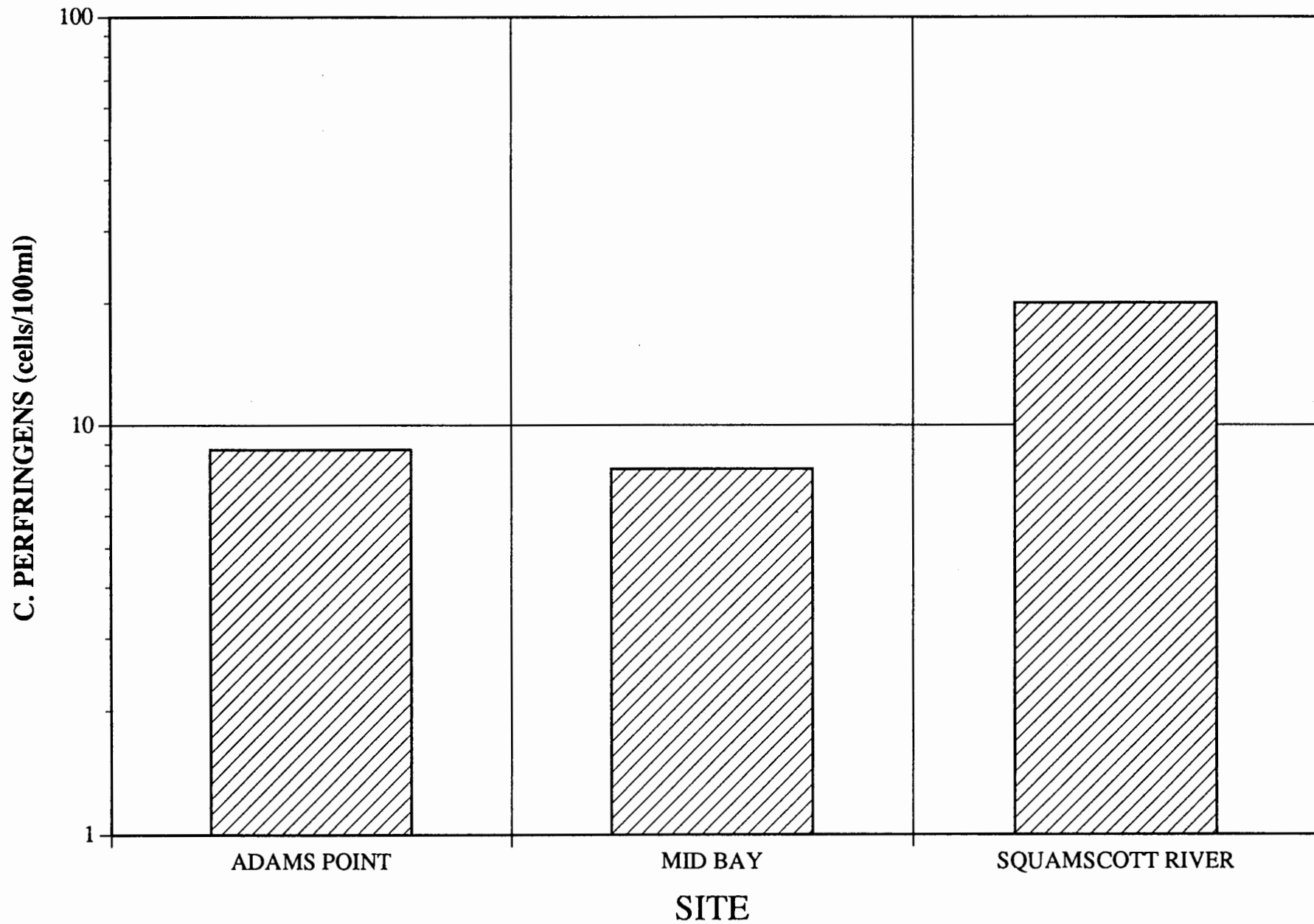


Figure 15. Geometric mean *C. perfringens* concentrations at low tide
6/91 to 8/92.

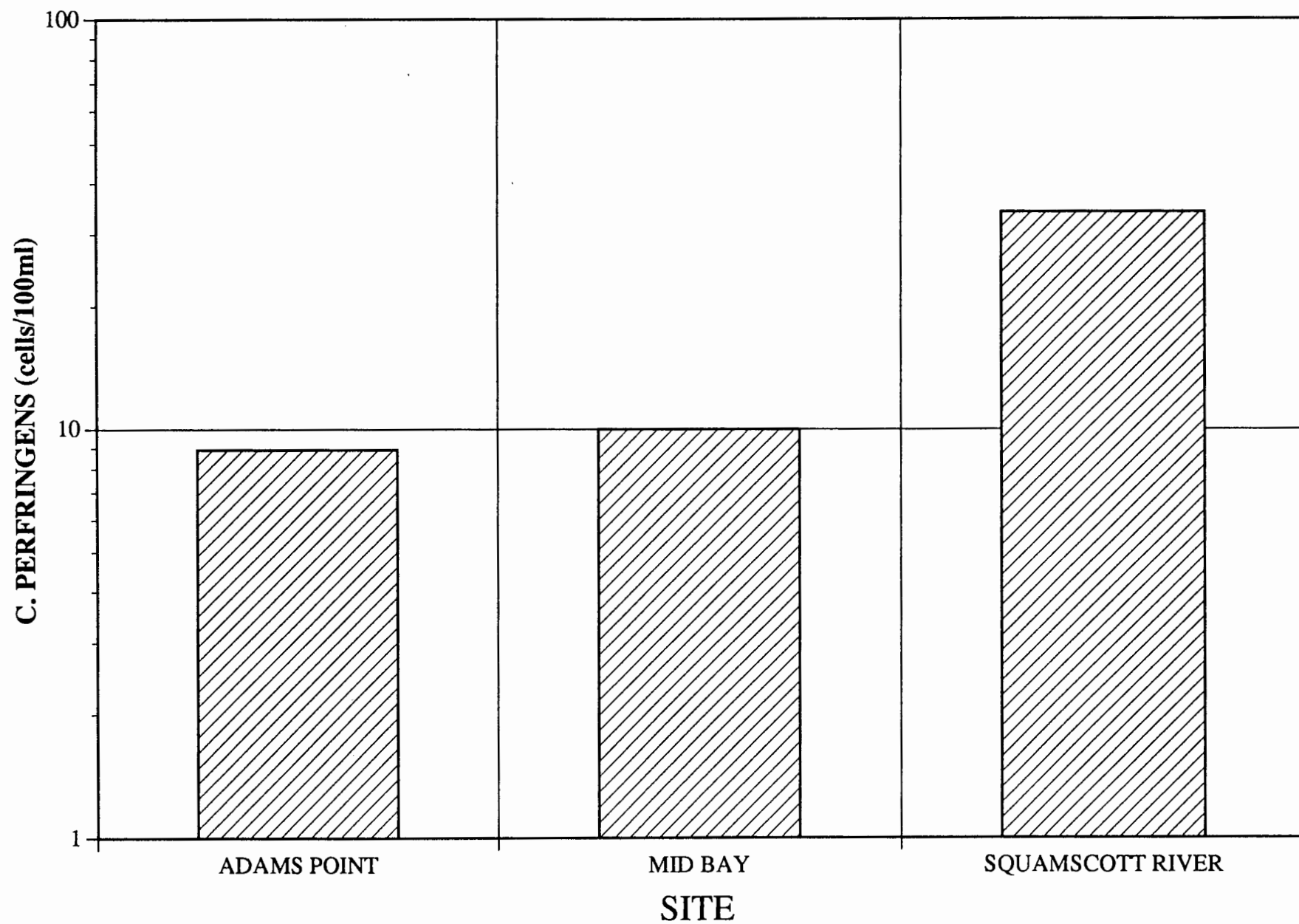


FIGURE 16
 TSS CONCENTRATIONS IN GREAT BAY
 PUMP SAMPLES - LOW TIDE - SURFACE

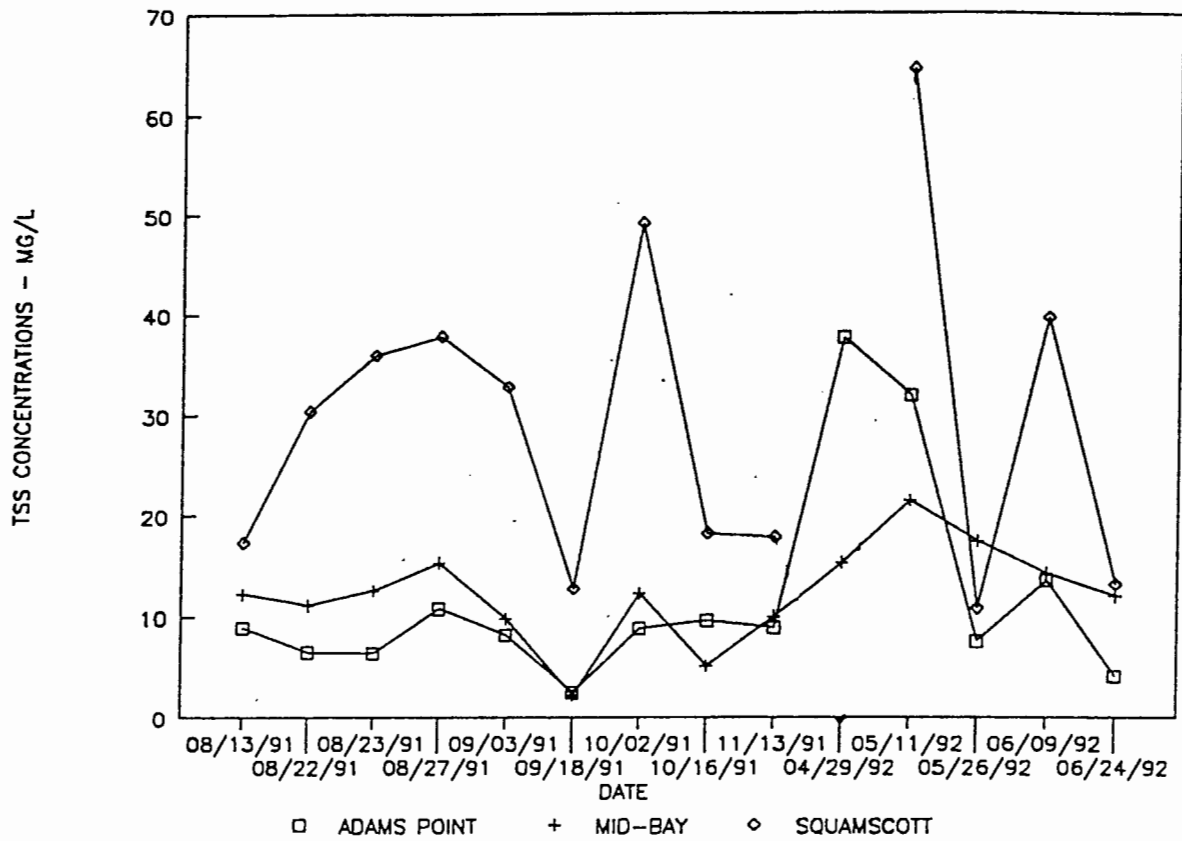


FIGURE 17
 TSS CONCENTRATIONS IN GREAT BAY
 HAND SAMPLES - LOW TIDE - SURFACE

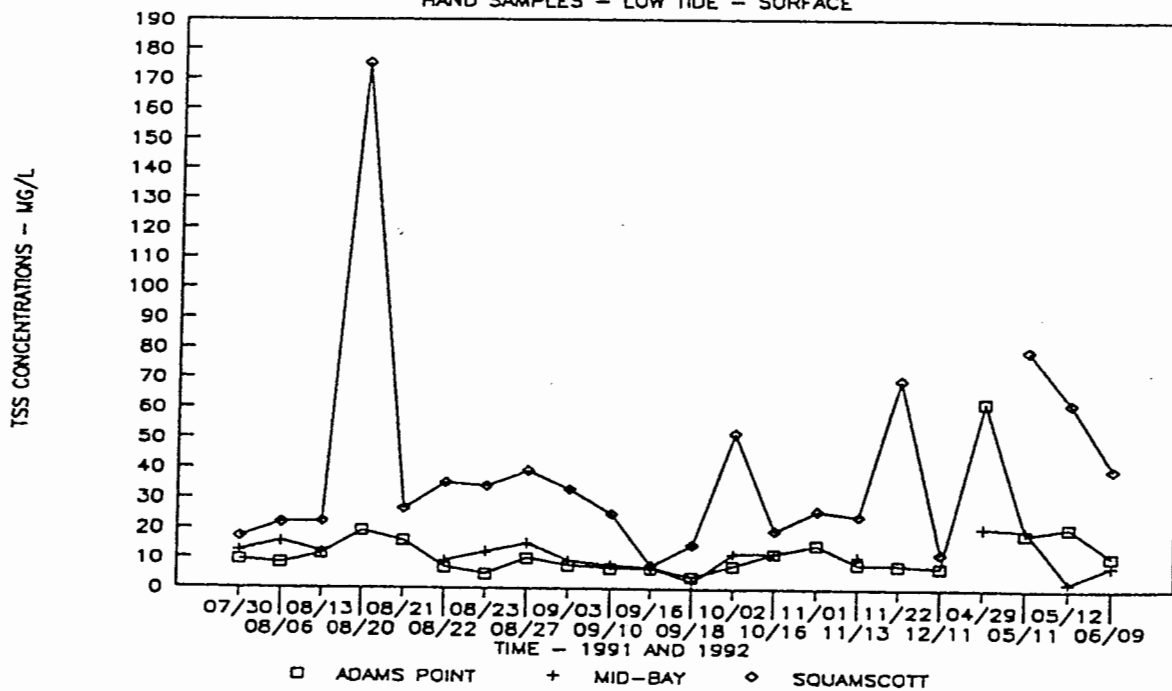


Figure 18. Enterococci concentrations at 3 sites in Great Bay before, during and after Hurrican Bob: August 19-20, 1991

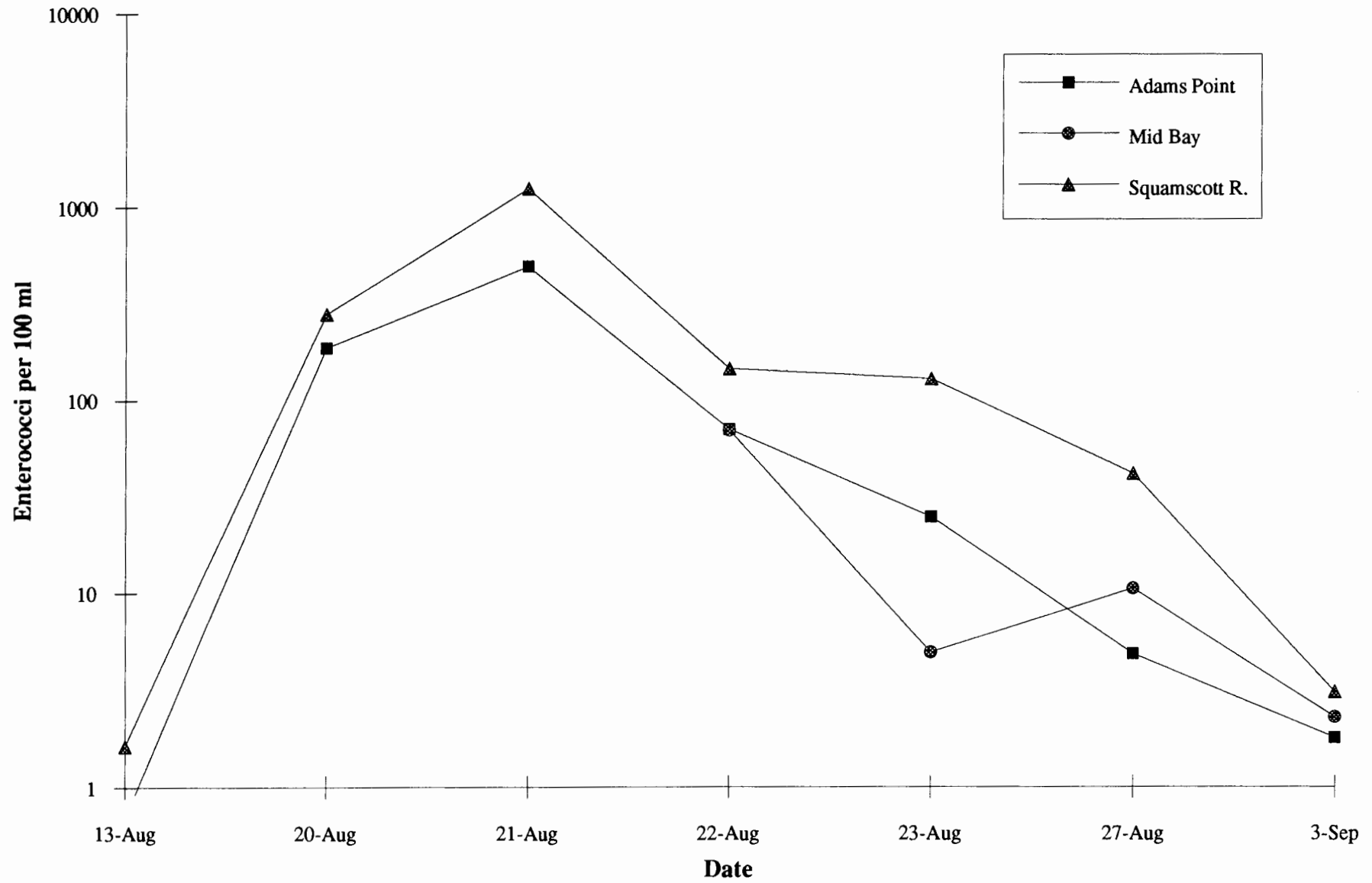


FIGURE 19
TSS - HURRICANE BOB

HAND SAMPLES - LOW TIDE - SURFACE

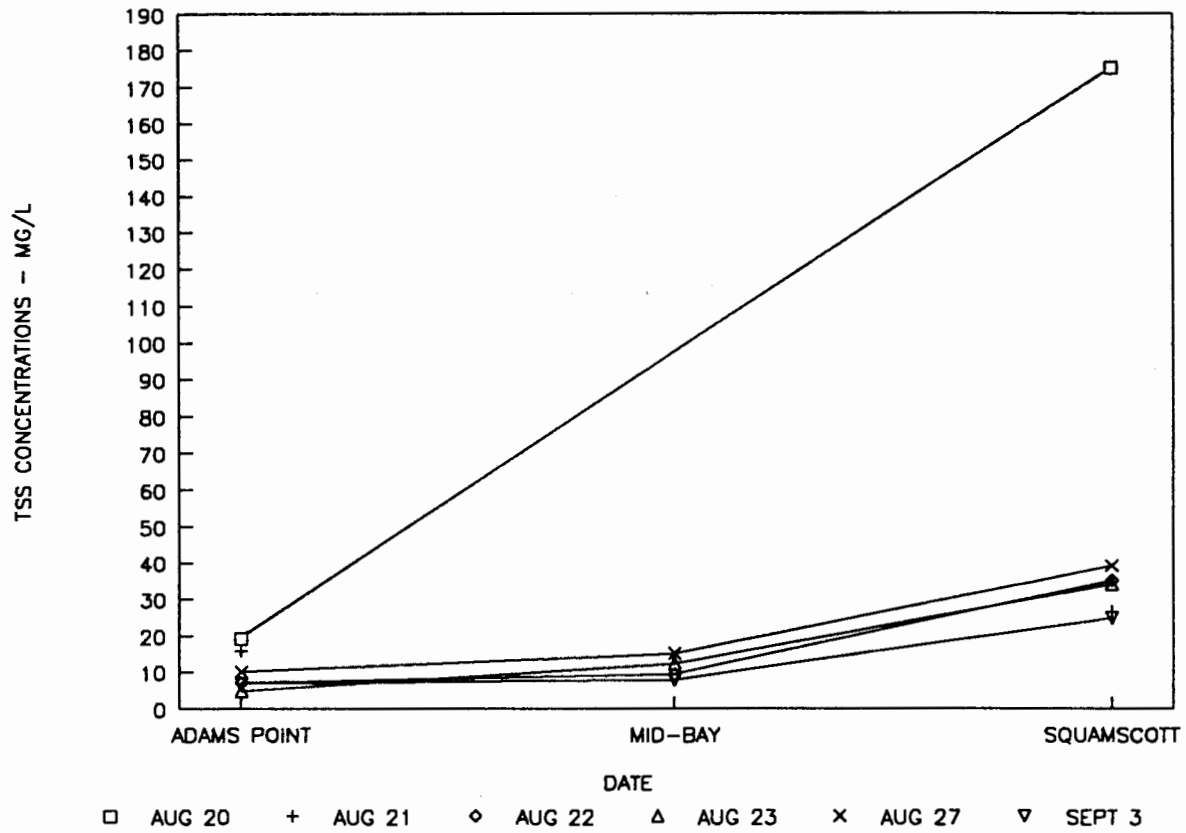


Figure 20. Relationship between enterococci and suspended sediments at Adams Point at high tide.

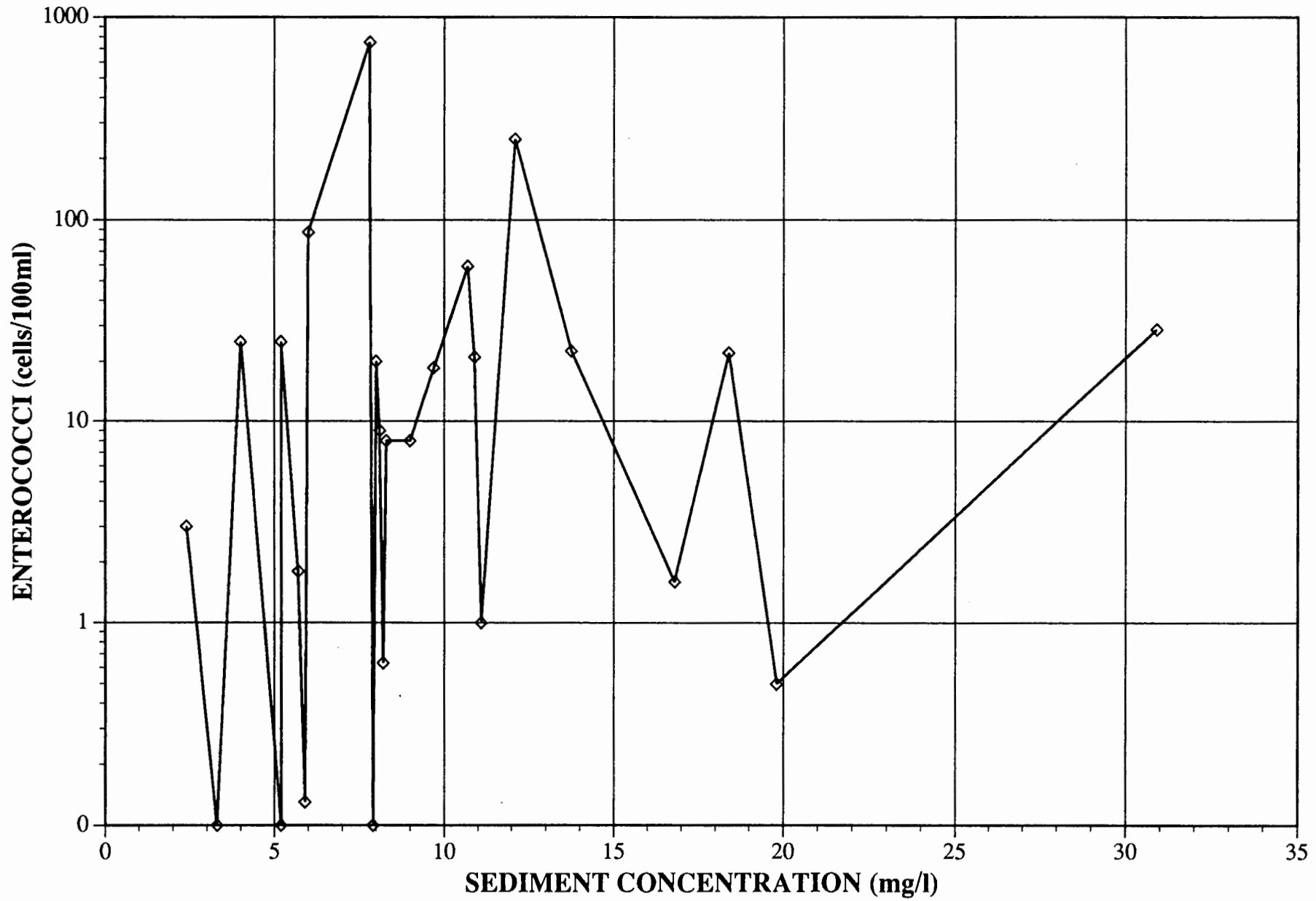


Figure 21. Relationship between enterococci and suspended sediments at Adams Point at low tide.

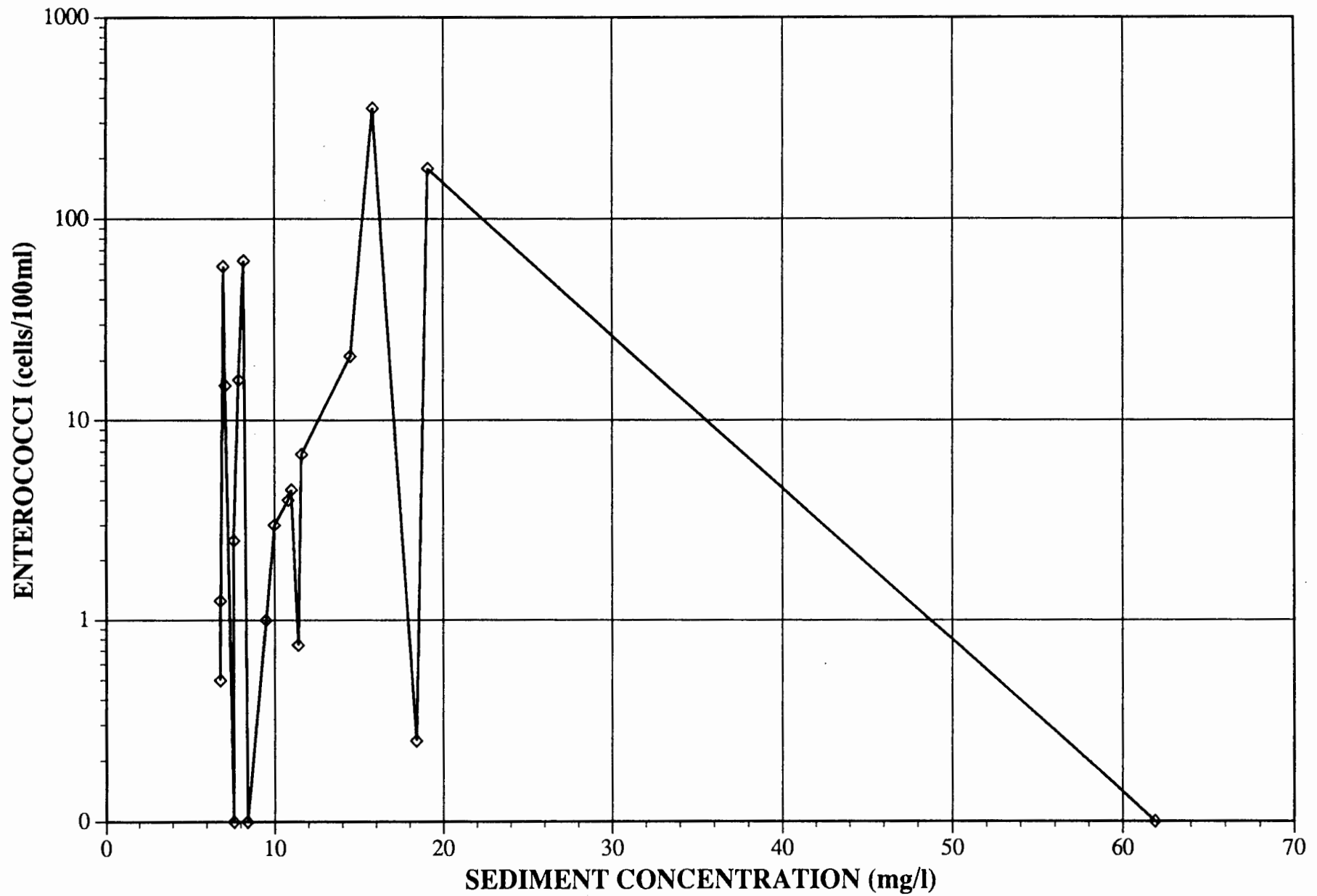


Figure 22. Relationship between enterococci and suspended sediments in Mid Bay at high tide.

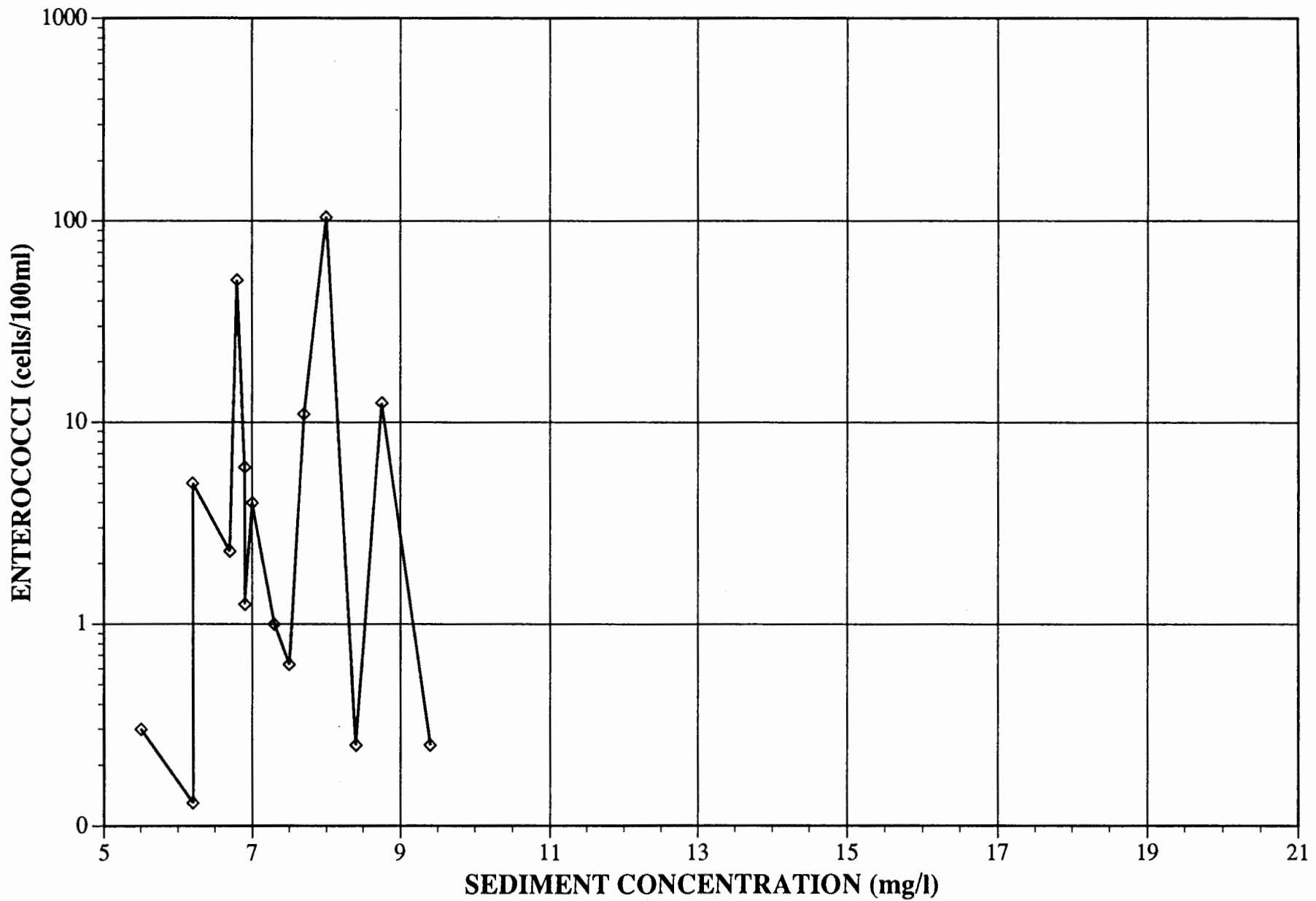


Figure 23. Relationship between enterococci and suspended sediments in Mid Bay at low tide.

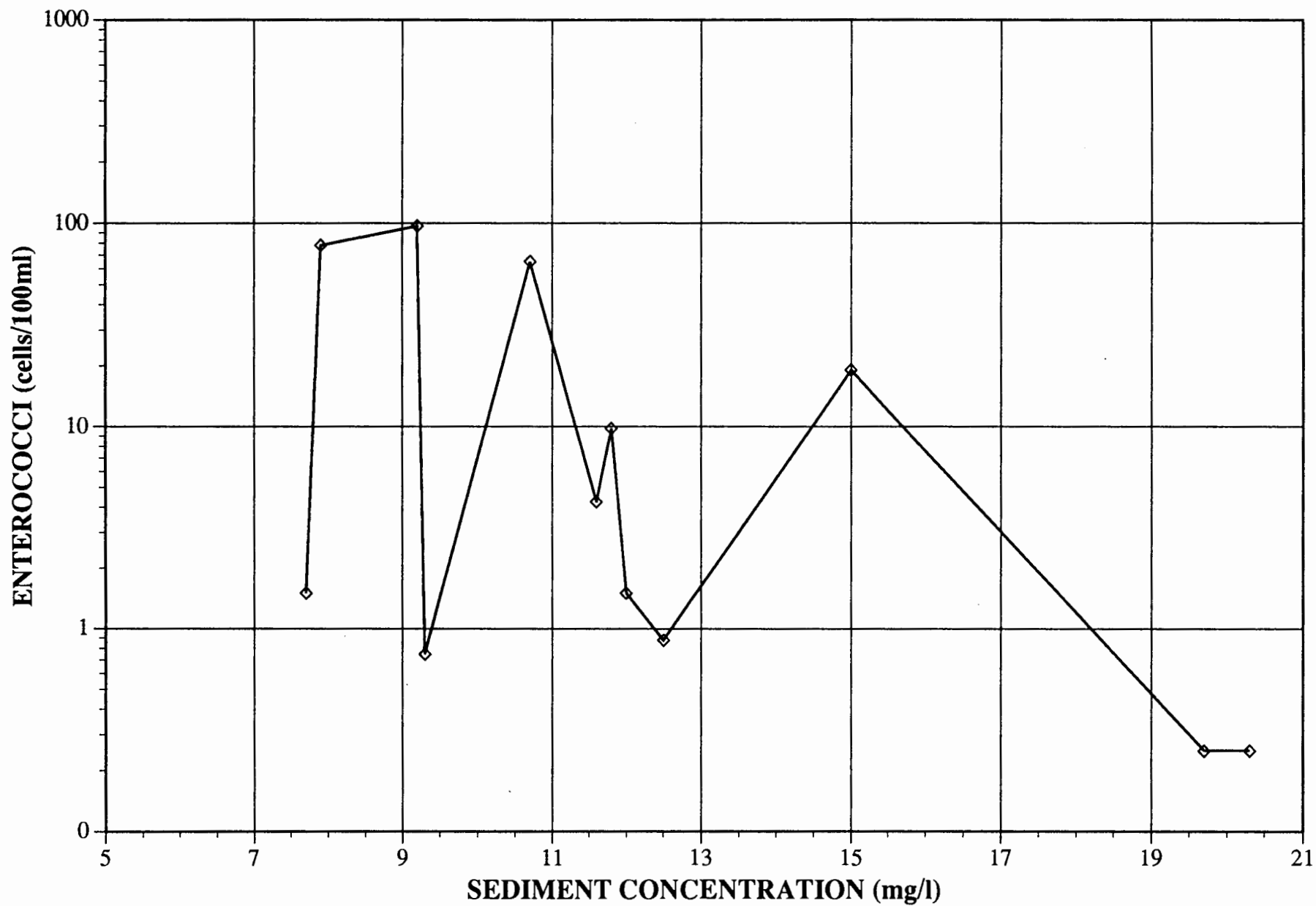


Figure 24. Relationship between enterococci and suspended sediments in the Squamscott River at high tide.

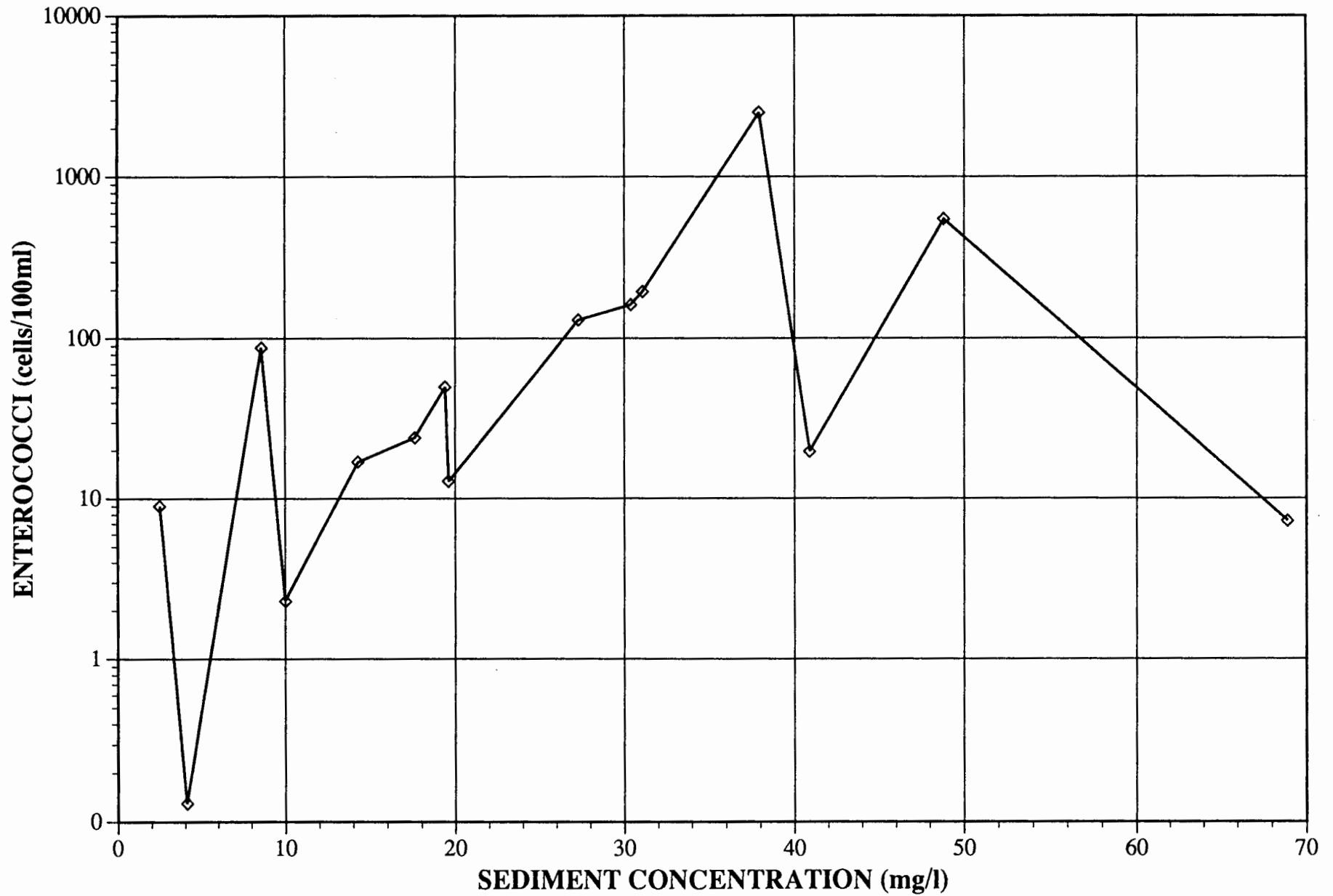


Figure 25. Relationship between enterococci and suspended sediments in the Squamscott River at low tide.

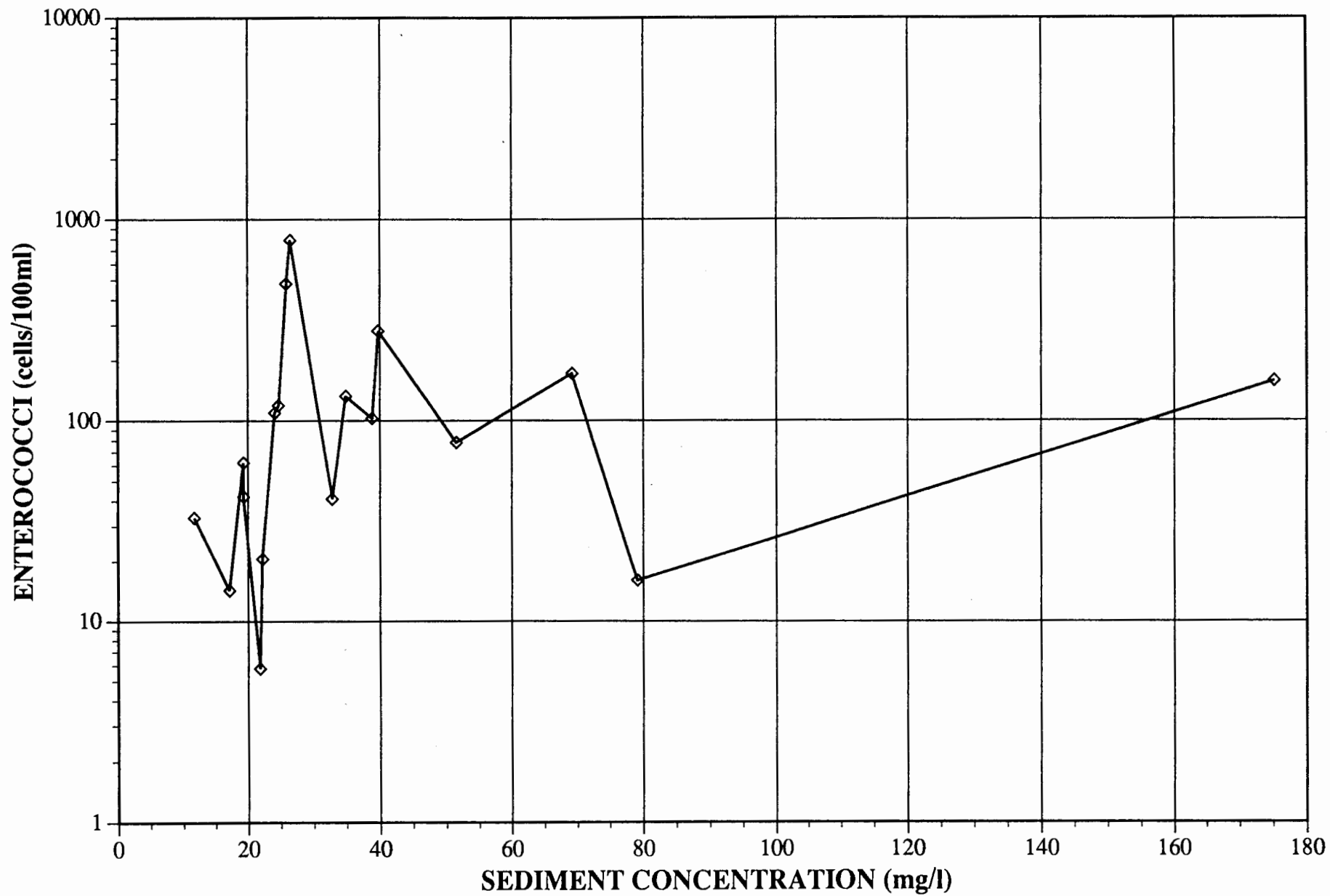


Figure 26. Relationship between fecal coliforms and suspended sediments in the Squamscott River at high tide.

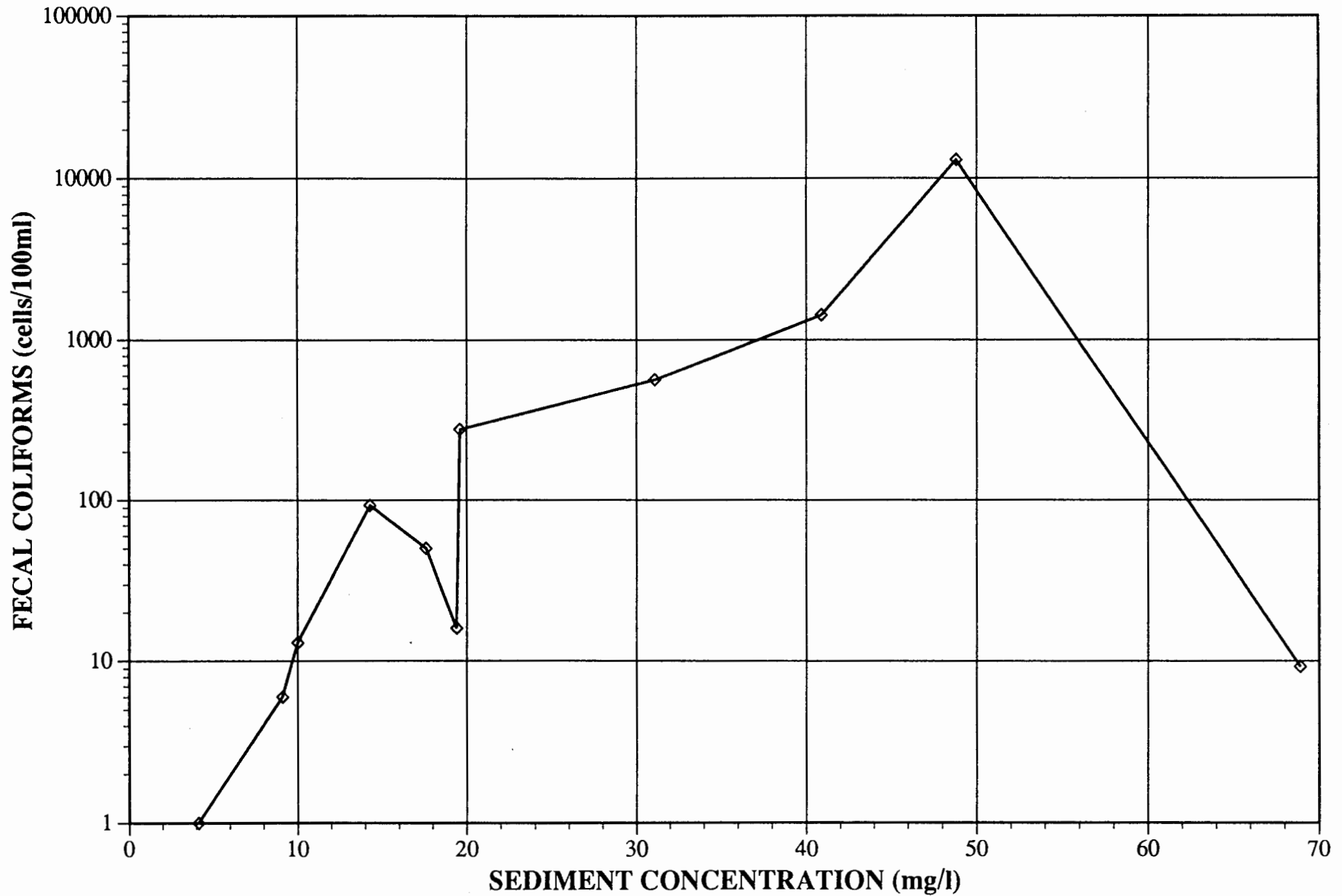


Figure 27. Relationship between fecal coliforms and suspended sediments in the Squamscott River at low tide.

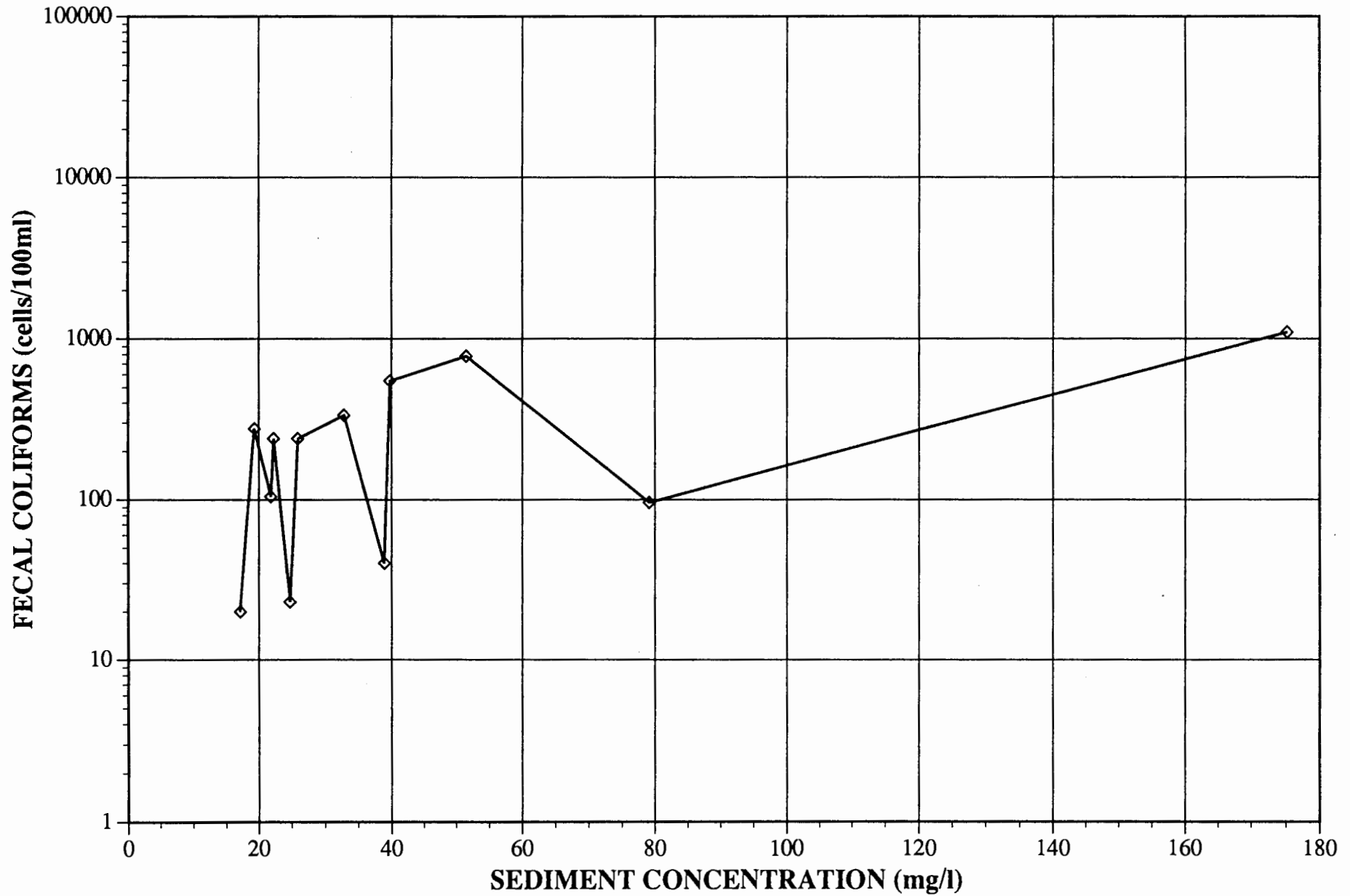


Figure 28. Relationship between *C. perfringens* and suspended sediments in the Squamscott River at high tide.

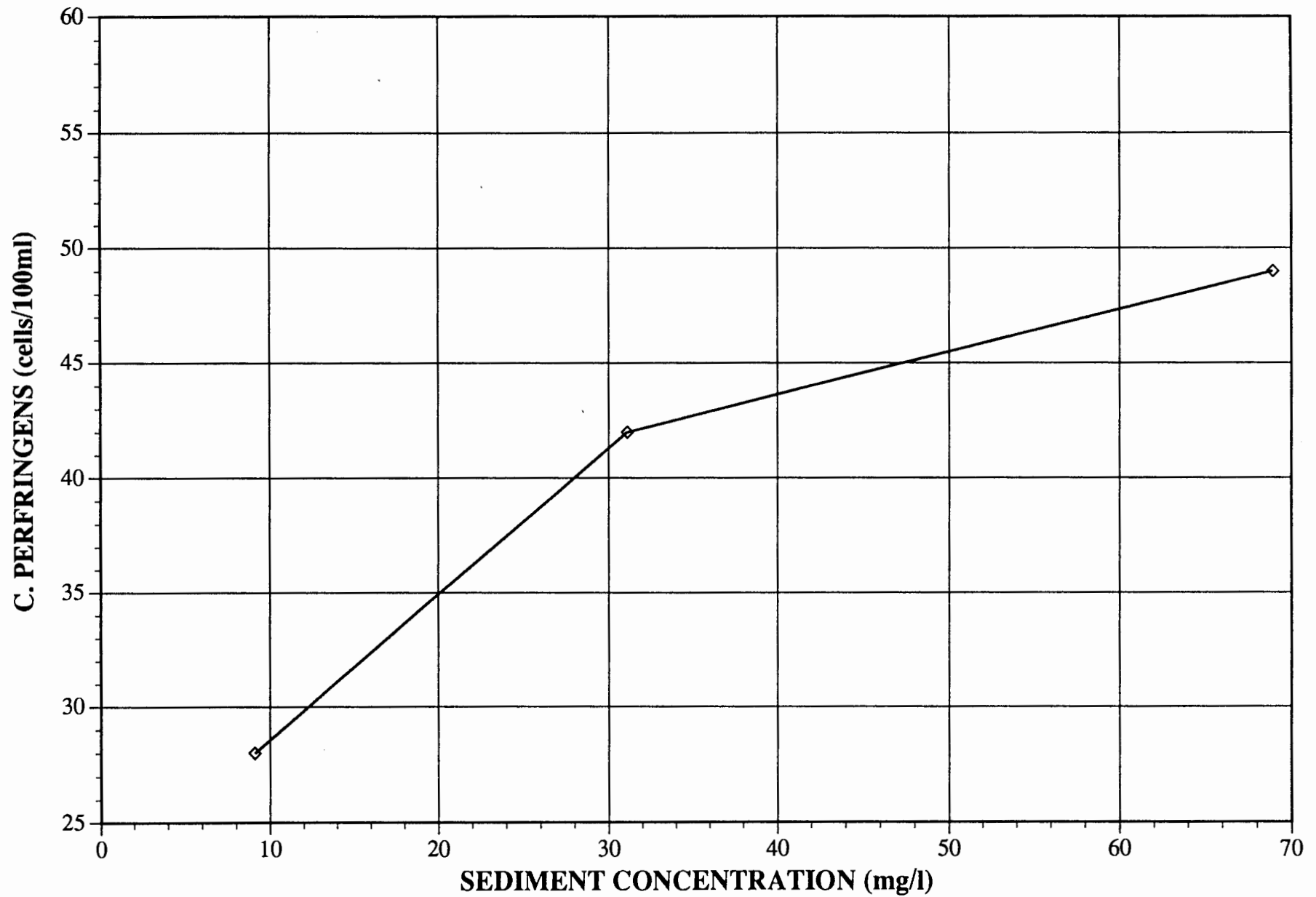


Figure 29. Relationship between *C. perfringens* and suspended sediments in the Squamscott River at low tide.

