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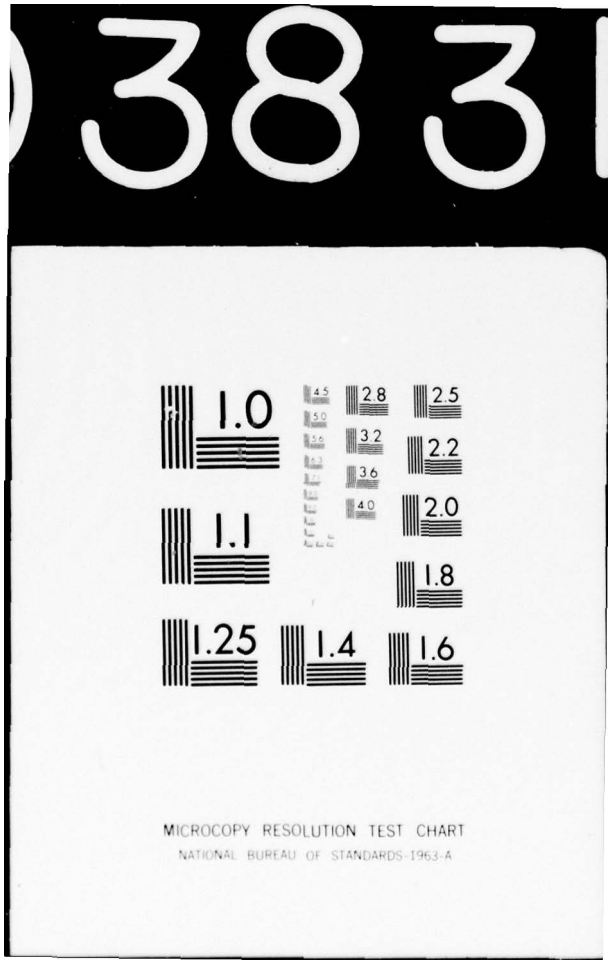
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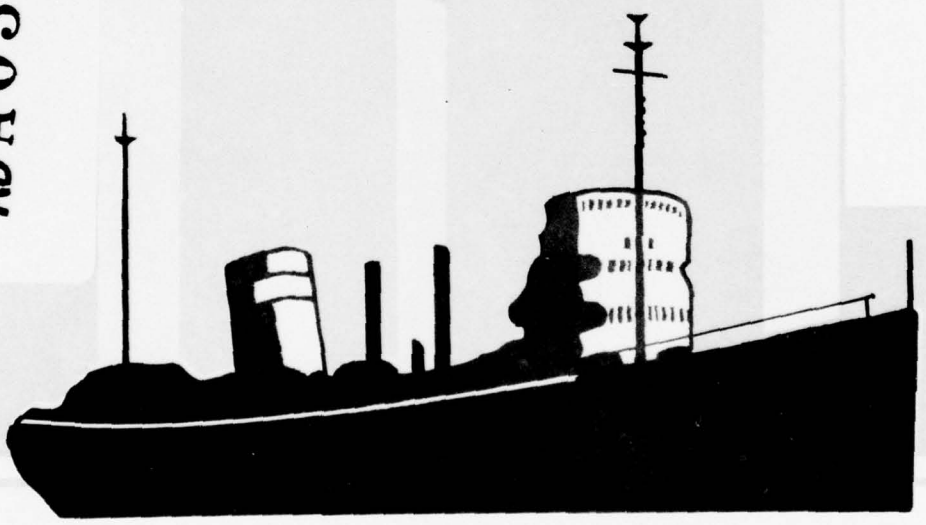

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The report presents the results of laboratory studies on the lethal impacts of suspended sediments on select macroinvertebrates and pelagic species. Initial sensitivity screening of San Francisco Bay species utilized commercial kaolin sediments. Four species were run with commercial bentonite varying and integrating levels of suspended solids, temperature, and dissolved oxygen at constant salinity. The ability of each species to survive burial was also studied.

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FOREWORD

In April 1972, the San Francisco District of the United States Army Corps of Engineers initiated a three and one-half year $3 million study to quantify the impact of dredging and dredged material disposal operations on the San Francisco Bay and Estuarine environment. The study is generating factual data, based on field and laboratory studies needed for the Federal, State and local regulatory agencies to evaluate present dredging policies and alternative disposal methods.

The study is set up to isolate the questions regarding the environmental impact of dredging operations and to provide answers at the earliest date. The study is organized to investigate (a) the factors associated with dredging and the present system of aquatic disposal in the Bay, (b) the condition of the pollutants (biogeochemical), (c) alternative disposal methods, and (d) dredging technology. The study elements are intended first, to identify the problems associated with dredging and disposal operations and, second, to address the identified problems in terms of mitigation and/or enhancement. The division into separate but inter-related study elements provides a greater degree of expertise and flexibility in the Study.

This report presents the findings of Appendix G, Physical Impact. The overall study will be the basis for preparation of a composite Environmental Impact Statement for Dredging Activities in San Francisco Bay System. A draft final report on the entire study is scheduled for completion in December 1975.

The following is an index of appendices to be published in the Dredge Disposal Study:

APPENDIX	REPORT	DATE PUBLISHED
	FINAL REPORT	
A	Main Ship Channel (San Francisco Bar)	June 1974
B	Pollutant Distribution	
C	Water Column (Water Column-Oxygen Sag)	
D	Biological Community	
E	Material Release	
F	Crystalline Matrix	July 1975
G	Physical Impact	July 1975
H	Pollutant Uptake	
I	Pollutant Availability	
J	Land Disposal	October 1974
K	Marsh Development	
L	Ocean Disposal	
M	Dredging Technology	

PREFACE

One of the four organizational areas of the San Francisco District's Dredge Disposal Study addresses the factors associated with dredging and the present system of aquatic disposal in the Bay. This area was segregated into physical, chemical and biological characteristics of importance for establishing the nature of the environmental system and the impact dredging and disposal operations exert. The physical studies were designed to investigate both phenomena in the water column and at the Bay floor. The biological studies were also delineated along these lines.

As part of the biological studies in this organizational area of investigation, laboratory experiments were performed to evaluate the effect of fine mineral particles in suspension and of cataclysmic deposition on estuarine macrofauna. The ramifications that dredging activities might have on the biological inhabitants of these two regimes (water column and benthic) have been discussed and judgement passed by many persons. However, quantitative information as to the magnitude of physical effects which could be expected under different levels of water column and benthal loading was wanting. As such, these experiments were undertaken concurrent with a field monitoring program to determine levels of sediment disturbance associated with dredging and disposal operations. The field monitoring program is presented in Appendix C, Water Column.

This investigation was performed using a commercially prepared clay to eliminate the complicating factors often associated with natural sediments because of *in situ* contamination with toxicants and pathogens. These contaminating materials in natural sediments would confuse the interpretation of physical as opposed to chemical impact events, i.e., whether the organisms are reacting to the suspended particulates or to the substances associated with those particulates.

The study of suspended solids effects was divided into four phases. The initial phase evaluated only the responses of several species to various levels of suspended solids, with other environmental factors held at non-stressful levels. This initial screening was used to select some of the more sensitive San Francisco Bay species for the subsequent experiments. The second phase consisted of a stepwise series of experiments to assess the influence of temperature on suspended solids tolerance. The third phase examined the influence of depressed dissolved oxygen concentration on the suspended solids tolerance of the test species. The final phase involved a multifactor experiment in which the solids loading, temperature and dissolved oxygen were varied simultaneously. This provided complex test conditions approximating those conditions found at a dredge site. The stepwise approach allowed determination of the most critical levels of each factor and aided in evaluating synergistic effects.

The ability of each species to survive burial was studied at the temperature that seemed most critical from the suspended solids tests. Animals not previously exposed to high suspended solids were buried under four depths of sediment to a maximum of 10 centimeters.

The research outlined above provides basic insights into the effects of suspended solids on macroinvertebrates and pelagic species. The response levels recorded will be integrated with the results of the Water Column Study (Appendix C) and the Dredging Technology Study (Appendix M) to provide an evaluation of the biological effect of the suspended solid levels created during dredging and disposal activities in San Francisco Bay. The research conducted thus far with commercial clays has provided the basis for additional research being funded by the Waterways Experiment Station using natural San Francisco Bay sediments.

EFFECTS OF SUSPENDED SOLIDS ON SAN FRANCISCO BAY ORGANISMS

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ABSTRACT

This research evaluated the impact of the presence of fine mineral particles in the water column on the macrofauna of San Francisco Bay. A unique laboratory facility providing large aquaria with open, once-through flow of water with the desired suspended solids concentrations was employed. The initial research determined the 200-hour LC_{10}, LC_{20} and LC_{50} of suspended kaolin for 18 species of fish and invertebrates. The more sensitive species, including Mytilus edulis, Crangon nigricauda, Morone saxatilis and Cymatogaster aggregata, were selected for study of the influence of temperature and dissolved oxygen on the 10-day LC_x of suspended bentonite, which approximated the fine sediments of San Francisco Bay in mineralogy and particle size distribution. Increasing suspended bentonite concentrations reduced the survival of all species, and tolerance decreased as dissolved oxygen content was lowered. Tolerance of the invertebrates to suspended bentonite varied inversely with temperature, while the survival of fish increased at higher temperatures. Changes in oxygen consumption of Mytilus edulis and Morone saxatilis were noted with increasing suspended bentonite concentrations. In burial experiments only Mytilus edulis and Synidotea laticauda had significant mortalities within 4 days after rapid deposition of up to 8 cm of bentonite. No species occurring primarily on muddy bottoms was found to be sensitive to high suspended bentonite concentrations. The sensitive species were all fish not intimately associated with the bottom, sandy bottom epifauna or fouling organisms, none of which normally encounter high suspended solids concentrations for extended periods of time. The results indicate that the impact of high suspended solids would be less in winter than in summer due to higher dissolved oxygen levels and lower temperatures.

CONCLUSIONS

1. The lethal concentration of suspended bentonite for 2-3 cm-long bay mussels Mytilus edulis was much lower than that of large mussels. Survival was reduced significantly by increasing suspended bentonite concentrations, with the effect exaggerated at summer temperatures. Survival was greater at saturated dissolved oxygen than at 5 ppm or 2 ppm, but little difference was apparent between the reduced levels. The short-term oxygen consumption of M. edulis in suspensions of bentonite was inversely correlated with concentration. The same experimental combinations of suspended bentonite, temperature and dissolved oxygen, eventually causing death in M. edulis, also resulted in loss of byssal attachments, but after much shorter exposure times. This may be an early and sensitive indicator of effective death. Mussels which became detached and fell to the bottom would be susceptible to covering by sedimentation, particularly near a dredging operation which created suspended solids high enough to cause detachment initially.

2. Under conditions of low temperature and saturated dissolved oxygen, survival of 3-5 cm sand shrimp Crangon nigricauda was high, even in high concentrations of suspended bentonite. Survival was reduced by summer temperature, even at saturated oxygen levels. Decrease in dissolved oxygen from saturation to 5 ppm dramatically reduced the tolerance to suspended bentonite.

3. Fingerling striped bass Morone saxatilis were killed at lower suspended

bentonite concentrations than any of the invertebrates tested. Survival varied inversely with suspended bentonite concentration and directly with dissolved oxygen and temperature. These factors were shown to interact in a complex, non-additive manner to reduce survival.

4. The most sensitive of the test organisms to suspended bentonite were 6-8 cm shiner perch Cymatogaster aggregata. As with M. saxatilis, increasing suspended bentonite concentration and decreasing dissolved oxygen and temperature combined in a complex manner to reduce survival. The slightly lower mortality of both species of fish at higher temperature was in contrast to all the invertebrates.

5. In none of the experiments did the magnitude of the 10-day LC_{50}, LC_{20} or LC_{10} values bear any predictable mathematical relationship to one another. This illustrates the necessity for studying the tolerance of the most sensitive members of a population.

6. Tolerance to suspended bentonite seemed to be correlated with normal habitat of the organisms, but no phylogenetic correlations were apparent. No species living primarily in close association with mud bottoms was found to be sensitive. All sensitive test species were either invertebrates occurring predominantly on sandy bottoms or in fouling communities, or fish not intimately associated with the bottom. Many tolerant species also were found in such habitats.

7. The results indicate that the biological impact of high suspended solids would be less severe in winter than in summer. The typically higher dissolved oxygen levels would increase the survival ability of all species studied. Low temperatures would increase the suspended solids tolerance of the invertebrates, but slightly decrease that of the fish. However, this slight reduction would likely be offset by the increased tolerance at high dissolved oxygen levels.

8. The primary emphasis of this study was mortality of adult macrofauna. It cannot be overemphasized that low mortality of adults in 10 days does <u>not</u> imply the absence of ecologically significant effects. Reduced reproductive success, either in terms of spawning adults, eggs, larvae or juveniles, may be of greater ecological importance than the death of part of the existing population.

INTRODUCTION

An analysis of the biological impact of dredging operations requires information obtained from both field and laboratory studies. This report describes laboratory research designed to evaluate the impact of the presence in the water column of fine mineral particles on estuarine macrofauna, without including the complicating factors often associated with natural sediments. A secondary objective was to determine the ability of selected organisms to survive rapid sediment deposition such as would occur on a spoil site. These two aspects of the research are treated separately in the report.

The report of the effects of suspended particles includes work begun under a previous research program and reflects changes in experimental design as the project evolved. These changes were primarily in the duration of the exposures to elevated suspended solids and a shift from marine species to estuarine organisms from San Francisco Bay. Because the effects of suspended solids on the test species was unknown, death was selected as the measured response to set outer limits of tolerance and provide a framework within which future research can be conducted. Consequently, the experimental suspended solids concentrations were high enough to produce significant deaths during the test period. It is extremely important to stress that the fact that no deaths were observed in some cases does not imply there was no effect on the animals.

The preliminary research design included the screening of a variety of species to select the most sensitive. Later experiments

considered the effects of elevated suspended solids at various levels of temperature and dissolved oxygen on the six species chosen for intensive study.

The primary attempt to quantify sublethal responses to elevated suspended solids was by measurement of changes in oxygen consumption. It was felt that respiratory surfaces would be constantly exposed to the action of the particles and that any interference might affect gill function and thus help explain any observed deaths. The same species selected for mortality studies were used in oxygen consumption measurements.

The successful completion of this project is due to the combined efforts of many people. The concept and basic design of much of the experimental facility was contributed by Clyde Davis. Gary Leo did the electrical engineering and assembly for the aquarium temperature control and data monitoring systems and Stan Read developed calculator programs for data storage and analysis. Statistical advice on the treatment of mortality data was provided by Mark Hudas. Many of the analyses were performed by Patricia Merrill, who also helped with revision of the manuscript. Tom Ronan and Jim Rutherford conducted animal checks at extreme hours and made possible the collection of mortality data at frequent intervals. Special thanks are due Mr. John Ladd of the California Department of Fish and Game for providing the experimental striped bass fingerlings, and Mr. Thomas Wakeman, U.S. Army Corps of Engineers, San Francisco District, for many helpful comments and for providing the particle size analyses. Dr. Cadet Hand, the Director of Bodega Marine Laboratory, was instrumental in the

initiation of this research, and provided valuable guidance throughout the project. Several revisions of the text and tables were patiently typed by Lorraine Andrade.

BIOLOGICAL EFFECTS OF SUSPENDED SOLIDS

HISTORICAL BACKGROUND

Relatively few studies of the direct effects of elevated suspended solids concentrations on aquatic animals are available. Many of these have been concerned with freshwater fish, primarily trout. Both lethal and sublethal effects have been studied for periods of exposure ranging from a few hours to several months.

The effects of many kinds of particles have been studied, including a variety of processed clay minerals, diatomaceous earth, powdered chalk, incinerator ash, coal washings and glass shards. Several investigations have used "natural" sediments taken directly from aquatic deposits. Often this material is sized, dried and resuspended, thus altering its physical, chemical and biological properties, before it is used in experiments.

The different methodologies employed in previous experiments make comparison of results difficult. A variety of techniques have been used to keep particles suspended, including periodic stirring with subsequent settling, continuous stirring and mixing by aeration. Almost all laboratory work has been done in closed aquaria systems, requiring either short tests or frequent changing of the water with consequent handling of animals. Many different methods have been employed in data analysis, also contributing to the difficulty of comparing results.

Few previous studies have related animal responses to actual weight per volume concentration of particles in suspension. Most correlated response with turbidity, an optical property of water containing suspended material, even though it seems unlikely that the

light absorbing and scattering properties of suspended particles directly affect animals. The turbidity produced by particulate matter depends on, among other things, particle size, shape, mineralogy and color, and there is no predictable correlation between the turbidities produced by equal weight per volume concentrations of different materials. This matter has been discussed by Kunkle and Comer (1971), who showed that turbidity could be related to weight/volume concentration of particles only if all the particles were of a uniform nature and instruments were calibrated against weighed samples. Turbidity is expressed in Jackson Turbidity Units (JTU), or in earlier work as equivalent to that produced by the stated parts per million of a standard silica flour. The latter is most unfortunate since statements like "turbidity of 1000 ppm" are easily misinterpreted as indicating a measured weight per volume concentration of particles.

One of the most widely quoted laboratory studies of the biological effects of suspended solids was by Wallen (1951). He exposed 16 species of freshwater fish to suspended soil particles, chiefly montmorillonite. The aquaria were stirred periodically by hand with an undetermined amount of settling between stirrings. Suspended solids were measured optically immediately after stirring and expressed as ppm turbidity. Maximum turbidities used exceeded 225,000 ppm, which probably was not the equivalent of 225,000 mgm/l or 22.5% solids on a weight per volume basis. Wallen found that deaths usually occurred within two hours after stirring, or the fish survived until the next period of greatly increased suspended solids concentration. All dying fish exhibited similar behavior, first gulping air and surface water,

then swimming slowly on their sides at the surface and finally floating on their sides with only occasional opercular movement until death occurred. The opercular cavities of all dead fish were matted with particles, but there was no evidence of structural gill damage. Most fish survived 100,000 ppm turbidity for a week or more but died within two hours when this was increased to 175,000 - 200,000 ppm turbidity. However, these levels are maxima at the times of periodic stirring and give no indication of the time-concentration variations to which the fish were exposed. Wallen concluded that the direct effects of montmorillonite clay turbidity were not lethal to juvenile and adult freshwater fish at levels found in nature.

A study of San Francisco Bay fish was conducted by the U.S. Fish and Wildlife Service (1970). They used Bay sediments at levels of 500, 1500 and 2500 JTU in 65 liter aquaria stirred electrically with a propeller. Changes in weight and survival in terms of fish-days were observed for up to 42 days. The 1500 and 2500 JTU levels caused significant mortalities and weight loss of survivors for all species, even though there were great species specific differences in absolute tolerance. They concluded that turbidities substantially above 500 JTU could affect the viability of fish.

The most comprehensive study of the effects of suspended solids on estuarine fish was by Sherk, O'Conner, Neumann, Prince and Wood (1974). They studied the effects of natural sediments, fuller's earth, and kaolin on the mortality, hematology, gill histology and respiration of a variety of estuarine fish encompassing many ecological types. The mortality tests were conducted for 12 to 48 hours in a

static system at about 22°C - 25°C. Kaolin proved to be the least harmful material tested and fuller's earth the most harmful. The authors found that the range from LC_{10} to LC_{90} (the concentration lethal to the stated percent of the population) could be large or small with no relation to the magnitude of the LC_{50} value. The species tested were classified based on their 24 hour LC_{10} in suspensions of fuller's earth. Class I (tolerant, 24 hour LC_{10} > 10 gm/l)included the mummichog Fundulus heteroclitus, striped killifish F. majalis, spot Leiostomus xanthurus, oyster toadfish Opsanus tau, hogchoker Trinectes maculatus and cusk eel Rissola marginata. The preferred habitat of all these species is the sediment-water interface where suspended solids are naturally at a maximum. Class II (sensitive, 24 hour LC_{10} of 1 gm/l to 9.9 gm/l)included the white perch Morone americana, striped bass M. saxatilis, bay anchovy Anchoa mitchilli, menhaden Brevoortia tyrannus, croaker Micropogon undulatus and weakfish Cynoscion regalis. Class III (highly sensitive, 24 hour LC_{10} < 0.9 gm/l)included Atlantic silversides Menidia menidia, the only highly sensitive adult form tested, and juvenile bluefish Pomatomus saltatrix, menhaden B. tyrannus and white perch M. americana. Dead fish commonly had a dense packing of particles in and around the gills, but no significant gill hemorrhaging. The authors suggested that the smaller gill openings of juveniles may have trapped more particles and at the same time the higher metabolic rate demanded more oxygen than adult forms. They also suggested that suspended solids effects may be related to particle shape and angularity, especially during long exposures where extensive damage to gill tissues might result.

Sherk et al. (1974) also investigated changes in hematology associated with suspended solids. Toadfish and striped killifish exposed to less than 1.25 gm/l fuller's earth for five days both showed significant increases in hematocrit and the hogchoker also had an elevated red blood cell (RBC) count. All these species were very resistant to death from suspended solids, but showed signs of sublethal stresses at low suspended solids concentrations. White perch, classified as a sensitive species, exposed to 0.65 gm/l fuller's earth for five days had significantly increased hematocrit, hemoglobin and RBC count, while the whole blood osmolality remained unchanged. When white perch were tested in natural sediments, these parameters increased, then gradually returned to control levels, perhaps implying some sort of compensating mechanism. Blood changes associated with high suspended solids were generally similar to those of fish deprived of sufficient dissolved oxygen.

The effects of suspended Chesapeake Bay sediments on the eggs of several estuarine fish were studied by Schubel and Wang (1973). Adult yellow perch were unaffected by concentrations below 500 mgm/l, but this level retarded hatching by 6 to 12 hours. Hatching success of white perch and striped bass eggs was unaffected by high suspended solids but 100 mgm/l and 500 mgm/l retarded hatching significantly. The authors concluded that under natural conditions suspended sediment concentrations up to 500 mgm/l probably would have little effect on hatching success of eggs of these species.

A study of the effects of suspended sediment on the eggs and larvae of white perch and striped bass from upper Chesapeake Bay was

conducted by Morgan, Rasin and Noe (1973). They found no effect on white perch hatching success and detected a slowed development rate only above 1500 mgm/l, much higher than the comparable threshold of 100 mgm/l reported by Schubel and Wang (1973). Particle concentrations above 4000 mgm/l delayed hatching up to one day. The hatching success of striped bass eggs was not affected below 2300 mgm/l, although development rate was lowered by more than 1500 mgm/l, again much higher than the 100 mgm/l found by Schubel and Wang. The two-day LC_{50} for white perch larvae was 2679 mgm/l and for striped bass larvae was 3411 mgm/l.

Rogers (1969) investigated the effects on several marine fish species of short exposures to a variety of suspended particles, including diatomaceous earth, kaolin, incinerator fly ash, ground rock flour, powdered charcoal, pulverized glass and glass beads. In 24 hour experiments mortality of all species was found to increase with exposure time and temperature, and with increasing particle size and angularity. Aeration of the water increased survival, indicating the fish were otherwise suffering from oxygen deficiency. The author concluded that suspended solids may affect fish in two ways, either by coating and clogging gills, or through abrasion of the branchial epithelium by rough particles.

Rainbow trout Salmo gairdnerii exposed to low concentrations of suspended kaolin and diatomaceous earth by Herbert and Merkins (1961) suffered 50% mortality after 185 days in about 270 ppm (0.27 gm/l) of both materials. Diatomaceous earth had no effect on length or weight increases among survivors at any concentration. The authors found many

cases of much thickened gill epithelial cells and frequent fusing of lamellae tips. The onset of such damage varied widely among individuals, with some fish affected after 11 weeks and some in the same concentrations still appearing normal after eight months. The authors noted increased incidences of fin rot associated with suspended diatomaceous earth.

Southgate (1960) found a median survival time for rainbow trout of 11 days at 270 ppm diatomaceous earth, which contrasts sharply with the 185 days reported by Herbert and Merkins (1961). Both investigations found similar changes in gill histology and Southgate interpreted frequent "coughing" actions by the fish as evidence of irritation of the respiratory surfaces.

While most research has centered on physiological effects of high suspended solids, Heimstra, Damkot and Benson (1969) studied behavioral effects of river bottom silt on small largemouth bass Micropterus salmoides and green sunfish Lepomis cyanellus. Suspended material was reported in JTU, with maximum test turbidities not exceeding natural levels, leaving the fish visible to observers. No short term behavioral changes were noted, but exposures of up to 30 days reduced general activity significantly in the bass. Neither species showed increased attack behavior related to suspended particles, nor a decrease in feeding. Both species "coughed" much more in turbid water, producing a violent sweep of water over the gill lamellae, interpreted by the authors as an effort to free them from accumulated particles. They felt the general reduction in activity could reduce the ability to find food and increase the susceptibility to predation.

Loosanoff and Tommers (1948), working with the American oyster Crassostrea virginica, found that natural silt concentrations as low as 0.1 gm/l caused a 57% reduction of pumping rate and 4 gm/l reduced pumping 94%. Similar results were obtained with kaolin but reductions were not so great at each concentration. Oysters ingested particles even in high suspended solids concentrations, and discharged much pseudofeces. The authors concluded that oysters were very sensitive to the presence of suspended materials and that there was a correlation between increased concentration and decreased pumping. Loosanoff (1961) continued this work for longer exposure times and also studied recovery of survivors in clear water. After 48 hours of exposure to muddy water oysters returned to clear water did not immediately show normal shell movement and resume rapid pumping, the typical behavior after shorter exposures. The author suggested that longer exposures might have injured the delicate ciliary mechanisms of the gills and palps.

The effects of several types of particles on the eggs and larvae of the hard clam Mercenaria mercenaria were studied by Davis (1960). In suspensions of 2 gm/l silt only 39% of the eggs developed to straight hinge larvae, but nearly all these metamorphosed when returned to clear water. Half the larvae died in 0.5 gm/l kaolin after 12 days, and growth did not occur at this concentration. Silt produced little mortality at 4 gm/l after 12 days, but no growth occurred and Davis inferred these larvae could not have metamorphosed successfully in nature. Growth was greatly retarded by 1.5 to 2 gm/l silt, but some larvae did reach setting size in 12 days. Generally larger particles affected the eggs most, perhaps by physical impact. Growth and survival of larvae were

most affected by small particles similar in size to food organisms, which mechanically blocked the digestive tract when ingested.

Davis and Hidu (1969) studied the effects of silt and kaolin on eggs and larvae of oysters C. virginica and hard clams M. mercenaria. Oyster eggs were affected more by the relatively large silt particles, while clam eggs were affected most by the small kaolin particles. The authors found that 0.19 gm/l of silt significantly reduced the number of oyster larvae reaching the straight hinge stage, and at 1 gm/l only 3% developed normally. Growth of oyster larvae was reduced at 0.75 gm/l, none could be measured at 2 gm/l, and all larvae eventually died at 3 gm/l.

The effects of suspended bentonite on four bivalves, among them Tapes japonica and Mytilus edulis included in the present research, were studied by Chiba and Ohshima (1957). Concentrations of 1 gm/l did not reduce pumping rate in any of the species tested, and the pumping of M. edulis increased with increasing concentration.

One of the earliest studies of the effects of silt on bivalves was by Ellis (1936). Using 18 species of freshwater mussels he found that most died when permanently covered by 1/4 to 1 inch of deposited sediment. Laboratory tests showed that suspended silt interfered with feeding, as mussels in muddy water remained closed 75-90% of the time and those in clear water were closed less than 56% of the time. All dead mussels contained silt in the mantle cavity and sometimes in the gill chambers.

The effects of increased suspended solids on the slipper limpet Crepidula fornicata were investigated by Johnson (1971). Field studies

showed a significant decrease in growth rate in higher concentrations of suspended sediment. In the laboratory Johnson found that increasing concentrations of kaolin, diatomaceous earth and silt all depressed filtration rate as concentration increased. The sharpest decrease was with silt between 0.14 and 0.20 gm/l. The author concluded that the increased pseudofeces production with increasing suspended solids concentration interfered with normal feeding and at the same time required an energy expenditure, accounting for the reduced growth he found in more turbid waters.

A similar suggestion was made by Pratt and Campbell (1956) studying the hard clam M. mercenaria. They counted expulsions of pseudofeces by clams in coarse sand, fine sand and mud. During simultaneous 30 minute observations clams averaged 3.4 expulsions per minute in coarse sand, 9.5 expulsions per minute in fine sand and 53.4 expulsions per minute in mud. The authors concluded that a greater intake of fine particles resuspended from the mud bottom required more frequent cleaning of the filtering apparatus, thereby increasing energy requirements, reducing active feeding time and wasting utilizable food due to incomplete sorting. These factors were presumed to have contributed to the slower growth they found in the field in mud bottoms than in sandy bottoms. A similar suggestion was made by Peddicord (1973) in regard to lower growth and condition index of the brackish water clam Rangia cuneata in mud bottoms than in sand.

The responses of the scallop Placopecten magellanicus and the mahogany quahog Arctica islandica to suspended kaolin have been studied (Stone, Palmer and Chen, 1974). No structural gill damage was found

but there was a proliferation of mucous-secreting cells on the gill filaments. In cleaning the gills of kaolin both species sloughed off increased quantities of mucous, which seemed to be derived metabolically from energy reserves in the digestive tissue. These reserves are also utilized in P. magellanicus by the gonads in gametogenesis, implying possible reproductive effects of exposure to high suspended solids at certain times of the year. The presence of suspended kaolin reduced the filtering rate of both species, the effect being greatest between 0 gm/l and 0.5 gm/l.

Little work has been done on suspended solids effects on crustaceans. Reeve (1963) studied the feeding of adult brine shrimp Artemia salina in mixed suspensions of food cells and soil particles. He found no selection between nutritive and non-nutritive particles and showed maximum filtration rate to be independent of the nature of the particles.

The effects of "red mud", the fine-grained residue of the extraction of aluminum from bauxite, on nauplii, copepodids and adults of the marine copepod Calanus helgolandicus were studied by Paffenhofer (1972). In mixtures of algal cells and 10 mgm/l red mud the success of molts to copopodid stage V and to adult was significantly reduced. All animals ingested much mud and produced many fecal pellets containing very few plankton cells. Growth, especially of older animals, was delayed in the presence of the red mud. The movements of adults were more sluggish in the mud than in the controls and females had no fat droplets and showed no ovarian development in the mud.

Sherk et al. (1974) compared the uptake of radio-labeled algal

cells in mixtures of silicon dioxide, fuller's earth, and silt by the copepods Eurytemora affinis and Acartia tonsa. All mixtures reduced ingestion rate at 250 mgm/l or more. Silt had the more detrimental effect, and A. tonsa was the more sensitive species. Feeding was stimulated by concentrations less than 100 mgm/l, perhaps due to some relationship between slightly elevated solids and increased food supply in nature. A significant reduction in carbon uptake by the phytoplankters Monochrysis lutheri, Nannochloris sp. and Chlorella sp. was found in 100 mgm/l to 500 mgm/l of silicon dioxide, suggesting a reduced food availability for filter feeders in muddy conditions.

Only those field studies of the biological effects of high suspended solids most relevant to this project are discussed here. May (1973) found normal winter background suspended solids in Mobile Bay, Alabama, varied from 2 to 133 mgm/l, with a mean at the surface of 27 mgm/l and 33 mgm/l at the bottom. Dredge discharge plumes were much more concentrated at depth than at the surface, and exceeded background levels over distances of 3000 feet or less downcurrent from the discharge. There was often a density flow, usually two feet thick or less, of very high suspended solids concentration. This flow exceeded 20,000 mgm/l for 1600 feet from the discharge and areas up to 2000 by 2400 feet had concentrations exceeding 1000 mgm/l. In the density flow, which moved independently of wind or tides, dissolved oxygen values were depressed to 2 ppm at 24°C. May concluded that the only direct effect of channel and shell dredging effluents on water quality in open Alabama estuaries was to temporarily increase suspended solids over a relatively small area. Most of the material settled into a

density flow several inches thick which had low dissolved oxygen and probably covered and smothered some benthic organisms such as worms and small molluscs.

Mackin (1961) found turbidities from small pipeline dredges to fluctuate widely and rapidly. Turbidities created by shrimp trawlers exceeded those 300 feet downcurrent from most dredges he studied. He concluded that particles were not carried more than 1300 feet and beyond a few hundred feet turbidities did not exceed normal maxima. He found little effect of particulate matter on oysters C. virginica in the field and considered oxygen sags of significant magnitude to be unlikely.

Biggs (1970), studying a channel dredging operation in Chesapeake Bay, found that the suspended solids plume at 3 m depth had a maximum extent of 4-5 square kilometers (1.5-1.9 square miles). It never extended farther than one tidal excursion from the discharge pipe and was not detectable 1-2 hours after discharge ceased. Over most of the area suspended solids loads were within the range of the observed natural yearly extremes, but occurred at different seasons from the natural maxima.

The effects of this same dredging operation on the fish of upper Chesapeake Bay were studied by Ritchie (1970). He could demonstrate no decline in the commercial striped bass M. saxatilis catch attributable to dredging and overboard spoiling. He caged four species of fish near the spoil site, and although results were variable, did not consider spoiling to contribute to mortalities. In a microscopic study of the gills of 51 fish of 11 species before and after dredging

he found no cell thickening or fusion of lamellae. The author concluded the least probable damage to fish from shallow overboard spoiling in upper Chesapeake Bay would occur in late December, January and February when fewest species are present.

Herbert, Alabaster, Dart and Lloyd (1961) studied the effects of fine clay particles on rainbow trout (Salmo gairdnerii) and the benthos of trout streams. Trout densities were inversely related to suspended solids concentrations. Fry were absent in muddy areas, although they were plentiful in similar clear water areas. Thickening of gill lamellae was found in trout in the more turbid waters. Molluscs were absent in streams with high suspended solids, but otherwise the effect was to reduce benthic faunal abundance but not alter its composition.

The literature on the effects of suspended solids on aquatic animals has been reviewed by several authors. Cordone and Kelley (1961) surveyed the research in fresh water and concluded that adults could probably tolerate the normal extremes of suspended solids, but that deposition would kill eggs, larvae and insect fauna and alter the characteristics of the bottom. Wilber (1971) concluded that most filter feeders are not affected below a certain concentration but that higher solids concentrations interfere with the filtering mechanism. He also felt that a given suspended solids concentration might be more harmful in normally very clear water than in usually muddy water. Based on catches of various types of nets in clear and turbid water Wilber also speculated that the efficiency of sight feeders might be reduced as turbidity increased.

Sherk (1971) discussed the concept that each dredging site has inherent physical, chemical and biological limits beyond which significant effects will occur and that suspended and deposited sediment affects living systems in many different ways. Lethal and sublethal stresses over chronic exposures may affect any life history stage in terms of behavior, activity, or metabolic function and may eliminate certain species. A significant reduction in reproductive success, either to spawning adults or to eggs, larvae or juveniles, may be of greater ecological importance than the loss of part of the existing population.

In a later review Sherk (1972) pointed out that the response of organisms may not be due to suspended solids concentration, but perhaps to the number of particles in suspension, their densities, size distribution, shape, mineralogy, presence of organic matter and its form, metallic oxide coatings or sorptive properties of the particles. He felt the assessment of effects in each project should consider (1) the type of particles to be suspended, their transport, and the substratum changes produced; (2) the biological activity of the water column and sediment-water interface; (3) the introduction to the water column of particles with associated chemical or biological components; (4) the relationship between the properties of the suspended matter and the species of the project area, their requirements and repopulation dynamics.

In the summary of an extensive study of dredging effects in upper Chesapeake Bay, Cronin (1970) suggested the following guidelines for dredging operations: (1) In estuarine areas with valuable fish nursery grounds and benthos and plankton populations, large environmental

modifications need careful planning to minimize damage. (2) Disposal of fine sediments on flat bottoms can cover very large areas by fluid flow and later movement and a wide safety zone is needed to protect vital areas. Disposal into basins affects smaller areas, but these may be unique in themselves. (3) Dredging and spoiling will destroy significant numbers of benthos at any time of the year. Thus special care is required to protect critically important areas. If the new substratum is similar to the original bottom composition, reduced or eliminated benthos may recover to its original state in one or two years. (4) Since estuaries are widely considered to be vital nursery grounds for a variety of fish, special consideration should be given to fish eggs and larvae.

Cairns (1967) pointed out that all environmental conditions operative at the time of dredging must be considered. Organisms can often compensate for temporary stresses from one factor if this process is not complicated by other stressful conditions. He suggested regulatory standards for increased suspended solids based on natural variation and background levels at low flow conditions and differing to suit various types of waters. Cairns realized extensive monitoring would be required to set these standards intelligently, but believed the cost would be returned by maximizing non-harmful levels of usage.

LABORATORY FACILITIES

The laboratory facility for the present suspended solids research was a much modified and improved version of that described by Davis and Nudi (1971). Twenty-four aquaria were arranged in three sets

of eight, with a once-through flow of particles and water, which were constantly introduced in the desired proportions. The temperature, salinity and dissolved oxygen in the aquaria were also controlled and these parameters plus pH were automatically and continuously measured and the data stored on magnetic tape for later analysis. Temperature and salinity control was available in three 400-gallon animal holding tanks. To minimize leaching all surfaces exposed to seawater were of relatively inert materials; primarily PVC, titanium, 316 stainless steel, clean vinyl tubing or epoxy paint.

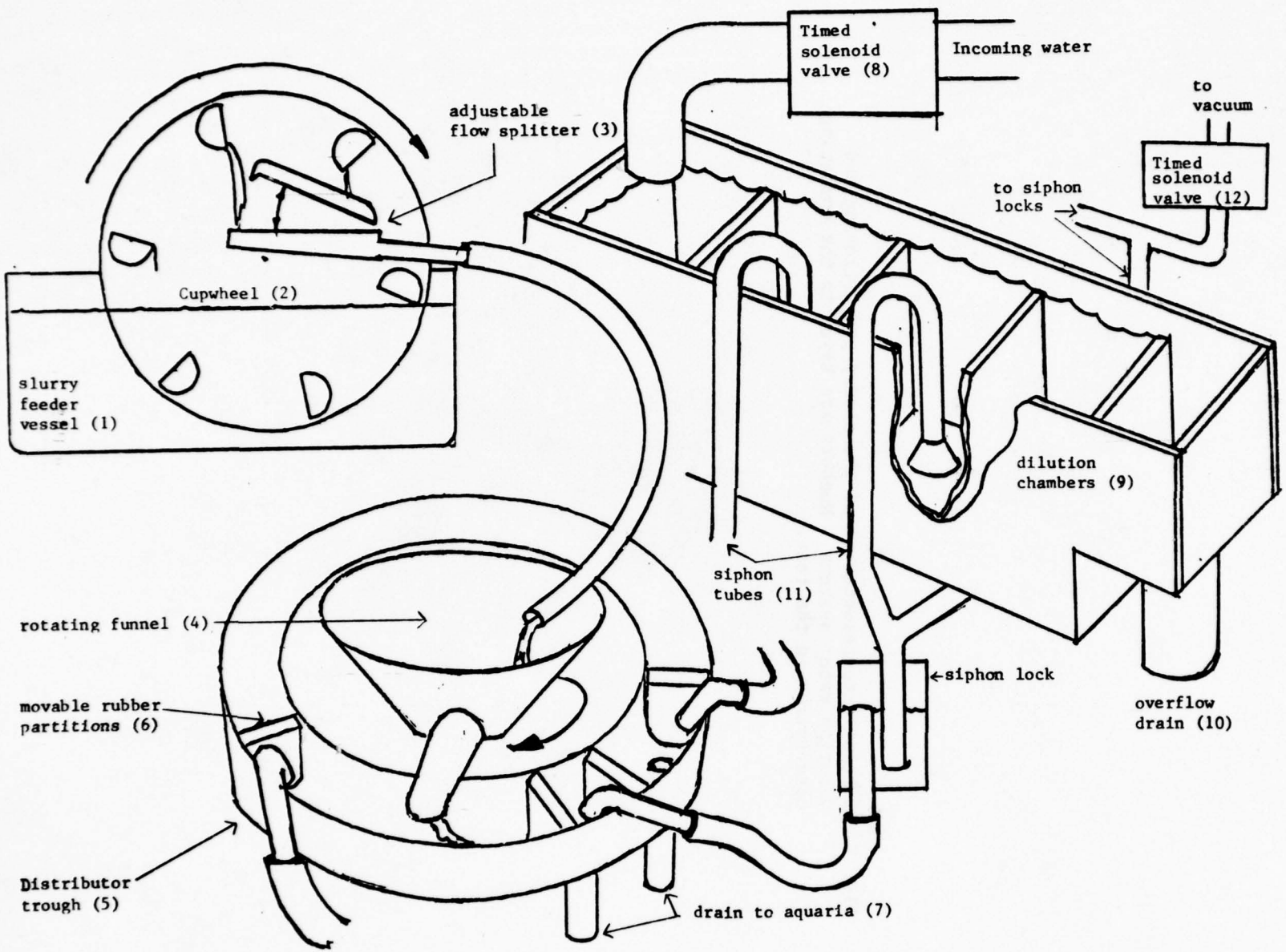
The experimental aquaria, constructed of fiberglass with an inert finish, were cylindrical with hemispherical bottoms and held approximately 84 liters of water. The particles were kept in suspension by a circulating pump for each aquarium which withdrew water six inches below the surface and re-introduced it through a disperser head in the center of the aquarium bottom. The water was forced out horizontally in all directions and flowed up along the rounded bottom and sides, constantly resuspending any material that tended to settle out. The water movement was sufficient to keep high concentrations of particles in suspension yet did not disturb small test organisms. Temperature control was maintained by passing the pumped circulating water through a heat exchanger before it was re-introduced to the aquaria through the dispersers.

The experimental animals were held in the aquaria in mesh baskets, preventing them from being drawn into the pumps, and allowing easy observation by gently lifting the baskets to the surface of the turbid water. Large animals were held in flat-bottomed 1/4-inch nylon

mesh baskets slightly smaller than the aquaria themselves. To prevent cannibalization of molting shrimp these organisms were tested in containers divided into individual cells by thin perforated PVC sheets. A mesh bottom and top were attached to allow free circulation of the water. Smaller organisms, such as amphipods and polychaetes, were held in baskets of nylon window screen with tight-fitting lids made from refrigerator food containers.

Mineral particles were introduced into the aquaria by a slurry proportioning system modified from the mineral dressing industry. A high shear impeller was used to mix the test material and water of the desired salinity to a concentration slightly above 25% suspended solids. This slurry was pumped into two 400-gallon polyethylene tanks with large stirrers and adjusted to exactly 25% on a weight/volume basis. From these reservoirs the slurry was pumped to three distribution systems which fed the three sets of eight aquaria. Each system, illustrated schematically in Fig. 1, consisted of a slurry feeder vessel (1) from which a cupwheel (2), driven at a constant speed by a gear motor, delivered slurry to a flow-splitter/receiver assembly (3). The opening in the splitter was adjustable to provide any required volume of slurry per unit of time. This predetermined amount of slurry flowed into a side-arm funnel (4) rotating at a constant speed in the center of a circular distributor trough (5) and was thus distributed evenly around the trough. Movable rubber partitions (6) divided the trough into sections whose size determined the amount of slurry each received. Each section had a drain (7) in the bottom from which the slurry flowed through a vinyl tube into the desired aquarium. Thus an

Fig. 1. Schematic representation of the slurry proportioning and dilution water systems. Numbers are keyed to the operating description in the text.


Timed solenoid valve (8)
Incoming water
to vacuum
adjustable flow splitter (3)
Timed solenoid valve (12)
to siphon locks
Cupwheel (2)
slurry feeder vessel (1)
dilution chambers (9)
siphon tubes (11)
rotating funnel (4)
siphon lock
movable rubber partitions (6)
overflow drain (10)
Distributor trough (5)
drain to aquaria (7)

individually controlled and constant amount of slurry was continuously introduced to each aquarium.

To produce the desired suspended solids concentrations, complementary volumes of water of the desired salinity were added to each aquarium with the slurry. This was accomplished with a modified form of the serial dilution apparatus of Mount and Brungs (1967). Water flowed through a solenoid valve (8) into a plexiglass trough divided into eight chambers (9), each of which filled and overflowed into the next and finally into a drain (10). The water valve then closed, and the excess water drained down to the top of the partitions. Each chamber had an adjustable siphon (11) running to the corresponding section of the distributor trough. The siphons were also connected to a common aspirator line with a solenoid valve (12). When the water chambers were full and stable, the aspirator valve was opened just long enough to start the siphons. These delivered the desired volume of water to compliment the amount of slurry already introduced into each section of the distributor trough, and thus into the aquaria. The clear water control tanks received their water directly from the siphon chambers, bypassing the slurry distributor trough. An identical system fed each of the three sets of eight aquaria. The amounts of slurry and water were controlled to produce the same turnover time in all the aquaria.

Salinity was controlled by mixing seawater from the laboratory system with fresh water from a spring-fed pond. The seawater recirculated through a swimming pool 20μ sand filter and passed through an ultraviolet sterilizer into a head tank. The fresh water was drawn

from mid-depth in the center of the pond and pumped directly into a second head tank. Valves regulated the flow of water from both tanks into a common pipe which delivered the controlled salinity water to the aquarium siphon chambers. All siphon chambers were filled through the same set of valves, producing similar salinities in all aquaria.

The use of two heat exchangers allowed experiments to be conducted at two different temperatures simultaneously. Both were of baffled counter-current design with the aquaria water passing through titanium or 316 stainless steel tubes 11 feet long. The controlling water in the heat exchangers was maintained at the proper temperature by an electrical feedback system controlling a proportioning valve and a set of hot and cold water supply valves. These controlled flow from a 55-gallon drum with an immersion heater, and a similar tank cooled by a 10-ton refrigeration unit. A thermistor in one of the aquaria provided a signal to the controller unit which actuated the appropriate set of valves, permitting hot or cold water to circulate through the heat exchanger body as needed to maintain a stable temperature in the aquaria. The proportioning valve allowed recirculation of the water in the heat exchanger body and bled in just enough of this new water to make the desired change. This design prevented large or rapid oscillation and minimized the load on the refrigeration unit.

The dissolved oxygen in the aquaria was normally maintained at saturation by the circulating pumps and saturated new water. However, oxygen levels could be lowered by stripping dissolved oxygen from the water with nitrogen gas. The pressure from a series of five nitrogen tanks in a cascade arrangement was maintained at 10 psi by a two-stage

regulator. Six low pressure regulators on a manifold further reduced this to 3 psi, each regulator feeding four needle valve flowmeters. These precisely controlled the amount of nitrogen entering each aquarium. The gas was introduced to the aquarium water on the pressure side of each circulating pump just before the disperser head, resulting in very fine nitrogen bubbles in the aquaria which effectively stripped the oxygen from the water.

A continuous record of temperature, conductivity, dissolved oxygen, pH and suspended solids concentration in each aquarium was provided by a remote automatic monitoring system. The pumped circulating water line of each aquarium had a branch line which diverted some water through a solenoid valve into a column containing sensors for the above parameters. An electronic scanner periodically energized each solenoid valve in turn, diverting a portion of the circulating water from each aquarium past the sensors. After three minutes a second scanner read the outputs from the instruments, the aquarium scanner closed that valve and moved onto the next aquarium. In this manner all five parameters were monitored in each aquarium every 72 minutes. The data were stored on magnetic tape for later analysis.

The temperature, conductivity, dissolved oxygen and pH monitors were part of a shipboard instrument package adapted for use in the flow-through sensor column. The temperature-sensing thermistors and conductivity induction cell were factory calibrated and verified against precise laboratory instruments. The dissolved oxygen meter was calibrated against Winkler titrations and the pH meter was calibrated using standard buffer solutions.

Suspended solids concentrations were monitored by surface scatter turbidimeters through which the water sample passed after leaving the sensor column. Two meters adjusted for sensitivity to different ranges of turbidity were used, the sample being directed through the proper one by a pair of solenoid valves. The optical instruments were calibrated against filtered and weighed samples from the aquaria so that a weight per volume measurement of suspended solids concentration could be obtained. Samples were pipetted from the aquaria, filtered through 0.45μ millipore filters, washed with distilled water, dried and weighed. A calibration cruve was developed by comparing the means of three replicate pipette samples to simultaneously obtained instrument outputs. Since stable, homogeneous and uniform particles were used in all experiments, optical instruments calibrated in this manner provided reliable measurements of concentration on a weight per volume basis.

Animals were held before testing in three 2' x 3' x 6' wooden tanks in which temperature and salinity were controlled. Water was mixed to the proper salinity by a pair of valves at the head tanks and introduced at a turnover rate of four hours. Temperature was controlled by continuously pumping the water through a stainless steel coil in either the hot or cold water reservoir of the aquaria temperature control system. A thermostat energizing an electrically operated three-way valve changed circulation from the heating to the cooling coil as necessary.

The oxygen consumption of the test species was measured at various levels of suspended solids using a modified Oceanography

International E/BOD Respirometer. This instrument consisted of a reaction vessel, manometric switch, electrolytic oxygen generator and an electronic readout unit. As dissolved oxygen was consumed, CO_2 was removed from the small air space in the reaction vessel by KOH pellets, producing a slight vacuum. This triggered a manometric switch and oxygen was generated electrolytically to replace that which had been removed. The current required to return the system to equilibrium was directly related to the amount of oxygen consumed. The design sensitivity of the instrument was 0.5 mg O_2. The reaction vessels used in the research with the mussels M. edulis were 3 liter round-bottomed pyrex chemical reaction kettles with lids. Mussels were placed in flat-bottomed mesh baskets in the vessels. For experiments with other species the volumes were reduced to one liter. A variable speed electric motor drove separate tubing pumps for each vessel, providing an airtight circulation system with very little heat input. The pump suction and discharge tubes, fitted through rubber stoppers in the reaction vessel lid, withdrew water at the bottom and re-injected it above the surface to increase mixing and gas exchange between the water and atmosphere. Temperature stability was provided by immersing the reaction vessels in a water bath which was circulated through one of the heat exchangers. The unique features of this system included the use of large reaction vessels, easy maintenance of solids in suspension, a continuous readout of oxygen consumption and the maintenance of dissolved oxygen near saturation so that the act of respiring did not continuously increase the stress on the animals.

EXPERIMENTAL METHODS

The criteria upon which the selection of experimental species was based changed somewhat as the research emphasis shifted early in the project from the nearshore marine environment to San Francisco Bay. The species chosen represented a variety of phylogenetic groups and feeding types, were abundant in San Francisco Bay, and were of major ecological importance. The first mortality experiments were conducted with kaolin, the simplest material tested, and the most sensitive species were selected for more detailed study with bentonite, a somewhat more complex material. A stepwise series of tests was conducted with bentonite, beginning with studies of lethal effects at two temperatures. This was followed by a set of experiments on the effects of suspended bentonite at two reduced dissolved oxygen levels while temperature was held constant, and finally by a multifactor experiment in which suspended bentonite, temperature and dissolved oxygen were varied simultaneously.

Most subtidal organisms, with the exception of striped bass Morone saxatilis and grass shrimp Palaemon macrodactylus, were collected in an otter trawl. Dipnets were used to collect a few species and all intertidal collecting was done by hand. The animals were transported to the laboratory in aerated ice chests in the water from which they were collected.

All strictly marine organisms were collected in Bodega Harbor and Bodega Bay, about 60 miles north of San Francisco Bay. This is a typical coastal marine environment with a seasonal temperature range of

about 8°C to 14°C, salinities usually near or above 30 °/oo, high dissolved oxygen and low background suspended solids levels compared to San Francisco Bay. Bodega Harbor is dredged about every six to eight years.

Collections of all estuarine organisms were from one of three areas in the San Francisco Bay system. In the South Bay near San Bruno Shoal Channel, which has been dredged regularly, salinity ranges seasonally from about 22 °/oo to 32 °/oo and temperatures are usually between 10°C and 20°C (Aplin, 1967). Suspended solids loads are higher than in the Bodega area but much lower than those characteristic of the northern portions of the Bay system. Collections were made at several sites in the Central Bay, including Southampton Shoal, Richardson Bay, Angel Island and Berkeley. Physical conditions here are similar to those in the South Bay. The most variable location from which collections were made was the Mare Island area, including northern San Pablo Bay and Carquinez Straits. Freshwater input is highly variable seasonally, resulting in nearly fresh water with high natural suspended solids loads in spring. The Mare Island Channel is dredged annually and the spoil is deposited at the west end of Carquinez Straits.

Striped bass M. saxatilis, the only species not collected at one of the above locations, were obtained from the hatchery of the California Department of Fish and Game. They were hatched in fresh water May 14, 1974, transported to the lab on August 14, 1974, in brackish water and held in the lab in water of 22 °/oo to 25 °/oo salinity. Saline water reduces parasitism and disease and has no apparent adverse effect on the fish (personal communication, Mr. John

Ladd, California Department of Fish and Game). Only one striped bass died in transport and mortality in the lab over a 2-1/2-month period was less than 1%.

Until used in experiments all other animals were kept in the laboratory holding tanks in water within ±3 °/oo of the salinity at which they had been collected. For experiments not involving temperature as a test variable, animals were held and tested within ±2°C of the temperature at which they had been collected. A period of acclimation was necessary for experiments conducted at other temperatures. When these studies were done collection temperatures were near the high test temperature, thus minimizing the need for warm acclimation. Animals were returned to the lab and divided into two groups, one of which was placed in a holding tank at collection temperature while the other was placed in a similar tank 2°C cooler. Temperature in the latter tank was stepped down 2°C every two days until the desired temperature was reached. Animals were held at this level for two or more days before the experiments began. Studies of temperature effects on suspended solids tolerance were conducted at 10°C and 18°C, approximately the normal seasonal extremes in San Francisco Bay. Experiments involving dissolved oxygen as a test variable were conducted at 5 ppm and 2 ppm. All other experiments were at saturated dissolved oxygen. There was no acclimation to the test suspended solids or dissolved oxygen concentrations since these could be abruptly changed by dredging activities.

For most mortality studies the experimental aquaria were arranged in three sets of eight, each set consisting of two clear water

control aquaria and six with progressively increasing concentrations of particles. During early experiments only two sets of aquaria were available and were arranged as replicates. For one experiment these 16 aquaria were arranged in four replicates of a control and three suspended solids concentrations. In the final multifactor experiment, each level of dissolved oxygen and temperature included a control and three suspended solids levels. The suspended solids concentrations always increased in an approximately logarithmic progression and nominally spanned one order of magnitude between the highest and lowest concentrations in a series. While this relationship was not precisely maintained, the concentrations were stable and close to the intended values. Turnover time in the aquaria was adjusted to four hours during the kaolin experiments and changed to six hours for the remaining bentonite studies.

The early screening tests were conducted for varying lengths of time, while the last screening tests and all bentonite experiments were continued for 10 days exposure time, which approximated the 12-day round-the-clock work periods in the San Francisco dredging schedule.

In order to make optimum use of the experimental facilities several species were tested together in the same aquaria. Only non-antagonistic species were tested simultaneously and they were isolated in separate baskets in the aquaria. The two species of fish, which were not aggressive toward each other, were combined in the large baskets. The species tested together in each experiment may be found in Table II of Appendices A, B, C and D.

To begin an experiment the aquaria were filled with water of the desired salinity and suspended solids concentrations, and the

temperature and dissolved oxygen were adjusted. The animals were then counted from the holding tanks into baskets and placed in the aquaria at the temperature and salinity at which they had been held.

The animals were observed three times per day at approximately eight-hour intervals throughout the experiments. The fish were checked only once daily during the suspended solids-dissolved oxygen experiments and multifactor experiments, and were fed adult brine shrimp after each observation. The death criterion was the absence of a detectable response to gentle probing of some sensitive part, usually the eye, siphon ends, mantle edge or mouth region. At each observation all dead organisms were measured to the nearest millimeter with the exception of the polychaetes, amphipods and isopods which often fragmented soon after death. During the experiments with bentonite, records were kept of shrimp that died in the process of molting, the number which had molted successfully, and the number of dead animals bearing eggs. During the multifactor experiments the number of mussels without byssal attachments was noted at each observation. Since the mussels were not attached at the beginning of the experiment, these observations included both those which did not attach themselves, and those which did not remain attached throughout the experiment.

After the bentonite tests the survivors were placed in clear, oxygen-saturated water of the same temperature and salinity to determine whether mortalities continued to occur during a post-exposure period after removal from high suspended solids concentrations. Observations were made daily for four days using the same death criteria as during the mortality experiments.

Studies of the effects of exposure to high suspended solids on oxygen consumption were conducted with previously unexposed individuals of the same species used in the mortality research. To begin a test the respirometer reaction vessels were filled with water of the proper salinity and suspended bentonite concentration and brought to 11°C or 18°C. The eight respirometer units were arranged in two replicates of a clear-water control and three increasing suspended bentonite concentrations.

Test animals, acclimated to the experimental conditions as described for the mortality experiments, were weighed in seawater and approximately equal numbers and biomasses were placed in all reaction vessels. The circulating pumps were then started and the organisms were allowed approximately two hours to adjust to their new surroundings and stabilize their oxygen consumption. After this period hourly observations of cumulative oxygen consumption were continued for 12 hours for M. edulis and for eight hours with the other invertebrates, while 30-minute observations were made for four hours with the fish. Five to eight experimental runs, depending on the variability of response, were conducted with each species.

Since some sedimentation occurred in the reaction vessels it was necessary to determine the amount still in suspension when the observations were made. Samples of liquid were withdrawn hourly, filtered through preweighed 0.45μ filter pads and washed with distilled water to remove residual salt. These were then dried at 100°C for 24 hours, reweighed, and the sediment weight converted to grams per liter. Suspended solids concentration was regressed on

time to estimate the amount remaining in suspension at any time.

DATA ANALYSIS PROCEDURES

The mortality data were analyzed for every eight-hour observation from the time of the first death until the end of the experiment. Mortalities were regressed on the corresponding suspended solids concentrations and the LC_{50}, LC_{20}, and LC_{10} estimated from the equation. The LC_{50}, LC_{20}, and LC_{10} values for all time intervals were then regressed on exposure time to estimate the time-concentration mortality response. By calculating the concentration lethal to 10%, 20% and 50% of the animals the effects on the most sensitive portions of the population were evaluated.

The observed mortalities in each test aquarium were adjusted for any deaths in the control aquaria by the method of Bliss (1935). The original number of animals exposed to each concentration was multiplied by the proportion of the original number alive in the control aquaria at each observation to give the adjusted original number in each test condition. The number observed alive in each treatment subtracted from the adjusted original number gave the number of deaths that could be attributed to the experimental variables. This number killed was divided by the adjusted original number to get the proportion of the test animals killed by the experimental variables.

The concentration of suspended solids in each aquarium at the time of every animal observation was determined for correlation with the mortalities. Data were recalled from the monitoring system storage

and the means of all concentrations recorded in every aquarium prior to each animal observation were calculated. In early tests when replication of experimental conditions was attempted, biological and suspended solids data were pooled in cases where an analysis of variance showed no difference in particle concentration between the intended replicates. Otherwise each aquarium was treated separately.

A programmable calculator was used to derive LC_x estimates for every observation time by the logit method of Berkson (1953). The mortality data adjusted for control tank deaths were converted to logits [defined as ln (P/1-P), where P is the proportion responding, or dying]. The logits of the mortalities in every aquarium were then regressed on the corresponding suspended solids concentrations to determine the relationship of mortality to concentration. Least-squares regressions were calculated using both arithmetic concentration values and natural logarithms of the concentrations and the equation having the highest coefficient of determination (r^2) was used. From this equation the concentration producing any logit, that is, any percent mortality, could be estimated. This process was used to derive LC_{50}, LC_{20}, and LC_{10} estimates for every observation time with each species.

The LC_x values were then regressed on exposure time to get estimates of the time-concentration mortality response. Least-squares regressions of the forms $y = a + bx$, $y = a + b\ln x$, $\ln y = a + bx$, $\ln y = a + b\ln x$, $\ln y = a + b(1/x)$ and $1/y = a + b(1/x)$ were fitted to the data points. These equations describe smooth lines that become more and more sharply curved in the order given. The equation that fit the data best, as indicated by the highest r^2, was used to describe

each set of data. The time-concentration mortality curves did not include any LC_x estimates higher than the highest suspended solids concentration tested, and began with the first estimate derived after x% mortality was actually reached.

The logit procedure, which does not consider 0% and 100% response, was unsuitable for analysis of the multifactor experiments since a partial response often occurred in only one of the three suspended solids concentrations. Therefore these data were presented as plots of percent survival vs. exposure time for each experimental condition. Quantitative analysis was by a 4 x 2 x 2 factorial analysis of variance (Sokal and Rohlf, 1969), considering four suspended solids concentrations, including the controls, two temperatures and two dissolved oxygen levels. The observations compared were lengths of time of survival of individual animals in a 240-hour experiment. A factorial analysis of variance provides a measure of the degree to which the experimental variables interacted to produce the observed results. The F values for the interaction terms measure only the non-additivity of effects of the particular factors, and provide no measure of the relative magnitudes of those effects. Therefore data producing significant interaction terms were re-analyzed so that valid mean contrasts could be made. This was done using a one-way analysis of variance considering each combination of levels of the interacting factors as separate treatments. Mean contrasts were by Tukey's W procedure (Sokal and Rohlf, 1969). The 95% confidence level was used in all statistical analyses unless otherwise stated.

The lengths of time individual M. edulis had byssal attachments

in the multifactor experiment were compared by an analysis of variance procedure analogous to that used for the mortality data.

In an effort to detect differential mortalities related to size, dead organisms were sorted by length class and an analysis of variance was used to compare the length of time of survival of the size classes. Data were compiled over all experimental conditions to provide large enough classes for meaningful analysis. An analysis of variance was also used to compare the length of survival of ovigerous and non-ovigerous C. nigricauda in the multifactor experiment.

The data on oxygen consumption at various suspended solids concentrations were converted to milligrams of oxygen consumed per gram of total weight for analysis. Data from reaction vessels in which a death occurred were excluded from the analysis. The relationship of oxygen consumption to exposure time was determined by a least-squares regression with multiple observations of Y (mg oxygen consumed per gm total weight) for every observation of x (time) as presented by Sokal and Rohlf (1969). Equations of arithmetic, semi-logarithmic and ln-ln form were fitted and the one having the highest r^2 was presented. Such an analysis was conducted for each set of experimental conditions. The oxygen consumption data were also examined using a factorial analysis of variance. The analysis of M. edulis considered the effects of time, suspended bentonite concentration and temperature on mg oxygen consumed per gm live weight. For the other species, which were tested successfully at only one temperature, a two-way factorial design involving suspended bentonite concentration and time was employed. In all cases the data analyzed were mg oxygen consumed per gm total weight

during consecutive time intervals. Significant interaction terms were re-analyzed using a one-way analysis of variance so that mean contrasts could be made.

EXPERIMENTAL RESULTS

Nature of the Particles

Commercially processed clay minerals were used as the experimental material throughout the project. The kaolin used in the early experiments was Hydrite Flat D (Georgia Kaolin Company), a hydrated aluminum silicate with a specific gravity of 2.58. Manufacturers specifications state the particles had a surface area of 18 m^2 per gram and were processed to retain less than 2% accessory minerals. The dry material had a median particle size of 4.5 μ with 10% of the particles finer than 0.5 μ equivalent spherical diameter and 10% coarser than 15 μ, making kaolin the most uniform material studied. Particles smaller than about 2 microns tended to exist as thin, flat hexagonal plates while larger particles occurred as stacks of these plates. Fig. 2 is an electron micrograph (5600x) illustrating the shape of kaolin particles.

Most of the experiments were conducted with bentonite (Clarolite T-60, Georgia Kaolin Company). This is a 3-layered aluminum silicate structure with a specific gravity of 2.59. The particles had a high cation exchange capacity and a surface area of 325 m^2/g. The dry material was air classified so that 90% of the particles were finer than 50 μ with a mean size of about 5 μ and about 35% finer than 2 μ (Fig. 3). As can be seen from the electron micrograph (1400x) in Fig.

Fig. 2. Electron micrograph (5600x) of the kaolin particles used in the initial experiments.

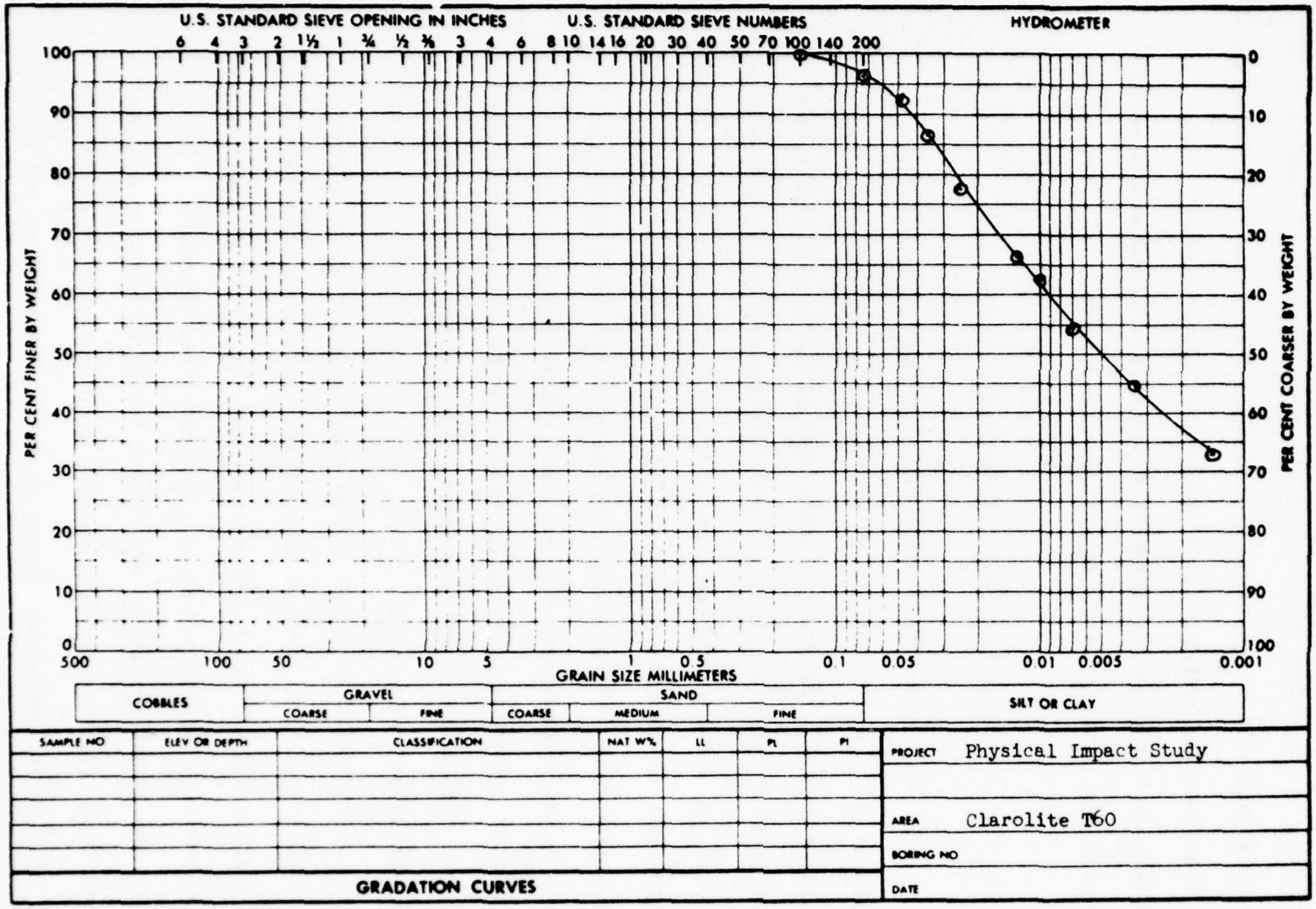


Fig. 3. Particle size distribution of the bentonite used in the suspended solids-temperature, suspended solids-dissolved oxygen and multifactor experiments.

4, the bentonite particles were jagged with irregular, abrasive surfaces. A comparison of the size distribution of the bentonite particles with the fine materials dredged at four sites in the northern San Francisco Bay System (Fig. 5) showed a high similarity. These natural fine sediments were chiefly montmorillonize and were mineralogically very similar to the experimental bentonite.

Lethal Effects of Suspended Kaolin

The conditions under which the experimental animals were collected, held in the laboratory and tested are presented in Appendix A. Table AI lists the species; date, method and site of collection; the length of time the animals were maintained in the lab before testing, and the salinity and temperature at which they were held. Table AII presents the conditions under which each species was tested, including duration of the tests; the salinity and dissolved oxygen common to all aquaria; and the suspended kaolin concentration, temperature and pH as individually controlled or monitored in each aquarium. The species tested simultaneously in the same aquaria are also identified in Table AII.

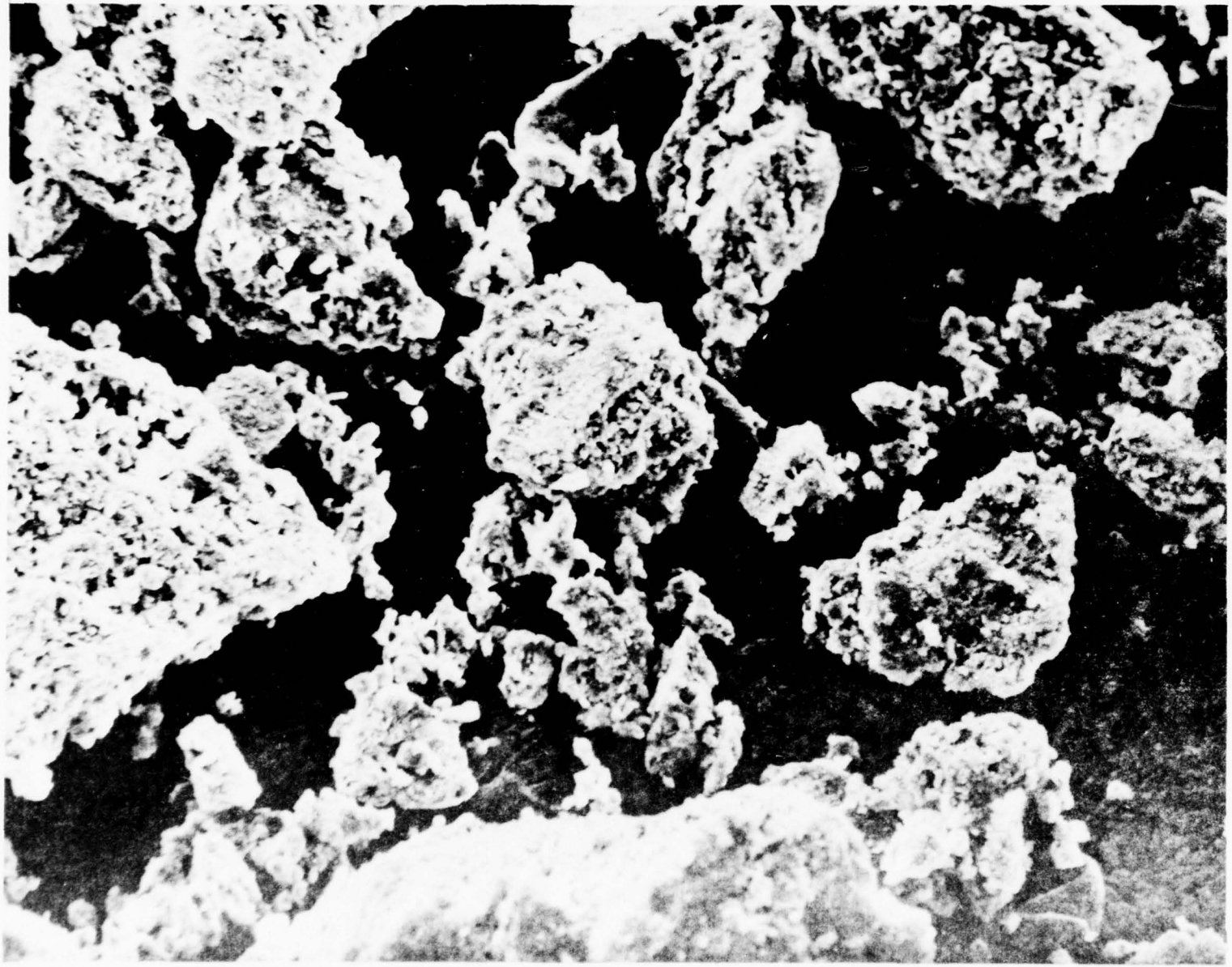
The experiments on the lethal effects of suspended kaolin indicated a very wide range of sensitivities among the species studied. Nine species did not reach 50% mortality in the length of time for which they were exposed and no LC_{50} estimates were possible. These species are listed in Table I with their length of exposure and the percent mortality observed in suspended kaolin concentrations of 100 gm/l. The purple sea urchin Strongylocentrotus purpuratus was not

TABLE I

Comparison of the mortalities of species relatively insensitive to suspended kaolin.

SPECIES	EXPOSURE TIME IN DAYS	% MORTALITY AT 100 gm/l
Strongylocentrotus purpuratus	9	0
Crangon franciscorum	5	25
Pagurus hirsutiusculus	12	0
Sphaeroma pentodon	12	0
Nassarius obsoletus	5	0
Tapes japonica	10	0
Mytilus edulis (2.5 cm)	5	10
Mytilus edulis (10 cm)	5	0
Mytilus edulis (10 cm)	11	10
Molgula manhattensis	12	9
Styela montereyensis	12	10

Fig. 4. Electron micrograph (1400x) of the bentonite particles used in the suspended solids-temperature, suspended solids-dissolved oxygen and suspended solids-temperature-dissolved oxygen experiments.



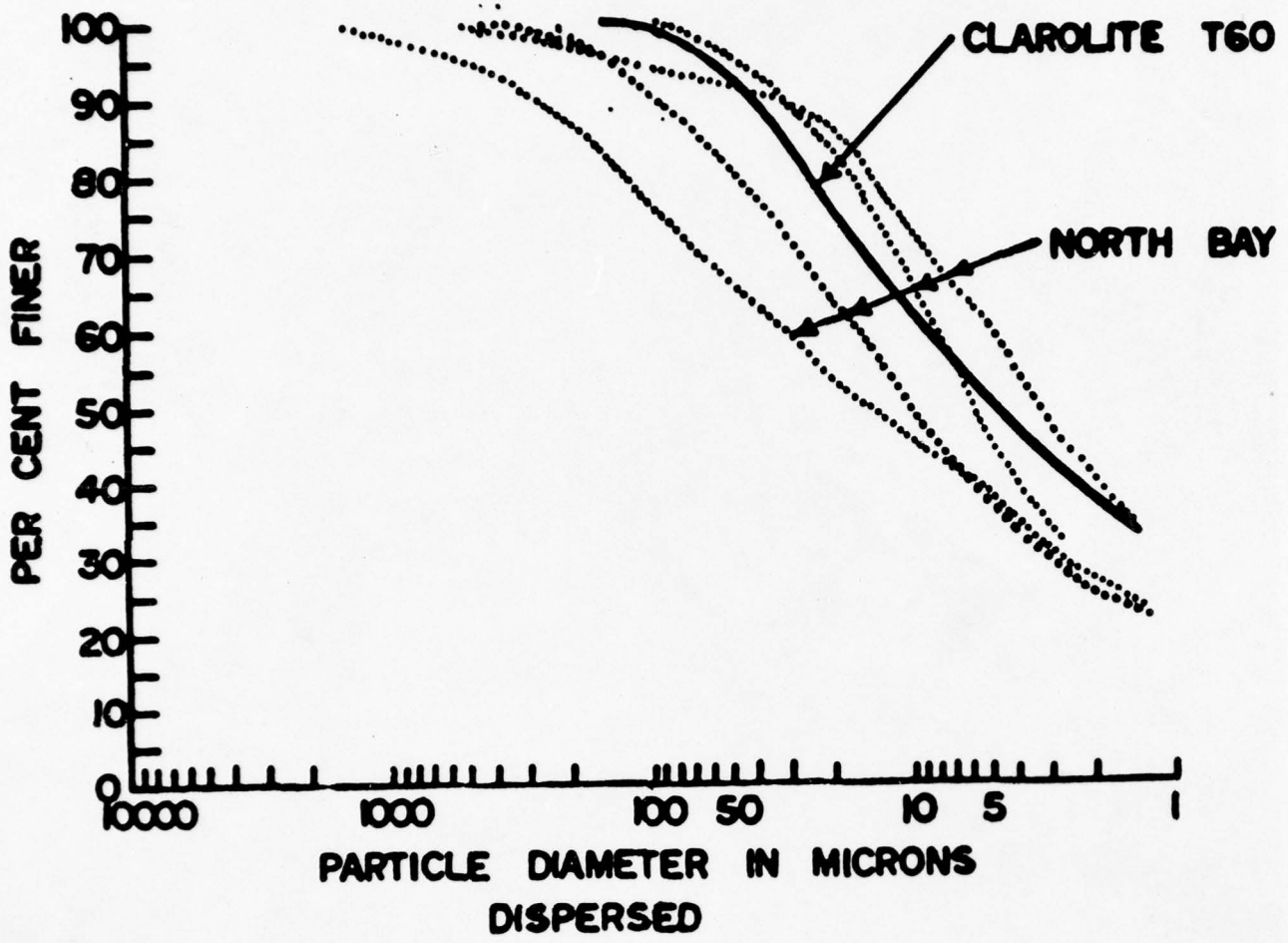


Fig. 5. Comparison of the particle size distributions of the experimental bentonite (Clarolite T-60) and natural fine sediments from four dredge sites in the northern San Francisco Bay system.

killed by suspended kaolin, but after nine days a pump failure in one aquarium allowed a coating of clay to settle on the animals, causing 100% mortality in a few hours even though they were not buried by the sedimentation. Blue mussels M. edulis 2.5 cm long reached 10% mortality after 5 days, while 10 cm long specimens had 0% mortality after the same time and did not reach 10% until 11 days of exposure. Two species of tunicates from San Francisco Bay, Molgula manhattensis and Styela montereyensis, had 12-day mortalities of about 10% at 100 gm/l.

A variety of species were more sensitive than the above. The raw data from the mortality tests with these species is summarized in Table II, showing the duration of exposure, the suspended kaolin concentration, original number of animals and number of deaths in each test condition. Table III presents the estimated 200-hour LC_x for each species, the equations from which the estimates were derived, and the r^2 values for those equations.

Figure 6 presents the LC_{50}, LC_{20} and LC_{10} curves for 10 cm long mussels Mytilus californianus exposed to suspended kaolin. This species, found along the open coastline, was more sensitive than the closely related M. edulis (Table I), usually found in bays and harbors. The estimated 200-hour LC_{50} for 10 cm M. californianus was 96 gm/l.

The time-concentration relationships for 50%, 20% and 10% mortality for the tunicate Ascidia ceratodes collected at Bodega are presented in Fig. 7. The experiment was terminated at 136 hours and the equation was extrapolated to derive a 200-hour LC_{50} estimate of 5 gm/l, making A. ceratodes one of the most sensitive species screened. This

TABLE II

Summary of raw data from mortality tests with suspended kaolin, showing species, length of exposure to test conditions and the suspended kaolin concentration, total number of deaths and original number of animals in each experimental condition.

Species	Hours of exposure	Suspended Kaolin Concentrations 1 gm/l	1 No. dead	1 Orig. no.	2 gm/l	2 No. dead	2 Orig. no.	3 gm/l	3 No. dead	3 Orig. no.	4 gm/l	4 No. dead	4 Orig. no.
Mytilus californianus	262	0	0	40	10	0	10	14	0	10	16	2	20
Ascidia ceratodes	136	0	3	80	10	7	20	12	4	20	15	10	20
Crangon nigromaculata	397	0	4	28	11	13	30	15	4	15	17	6	15
Crangon nigricauda	246	0	9	24	10	9	12	17	11	12	23	8	12
Palaemon macrodactylus	260	0	15	67	9	14	60	12	5	21	31	20	80
Cancer magister	240	0	7	19	11	6	21	14	5	19	26	9	19
Anisogammarus confervicolus	260	0	6	46	9	6	17	12	2	12	31	19	59
Neanthes succinea	260	0	7	28	9	8	20	31	13	28	102	20	20
Parophrys vetulus	238	0	0	30	10	0	15	17	0	15	23	1	15
Cymatogaster aggregata (Bodega Bay)	26	0	0	30	14	14	15	19	15	15	25	15	15
Cymatogaster aggregata (San Francisco Bay)	201	0	0	15	2.9	8	15	4.6	10	15	.6	0	12

Table II, continued

Suspended Kaolin Concentrations																				
5			6			7			8			9			10			11		
gm/l	No. dead	Orig. no.	gm/l	No. dead	Orig. no.	gm/l	No. dead	Orig. no.	gm/l	No. dead	Orig. no.	gm/l	No. dead	Orig. no.	gm/l	No. dead	Orig. no.	gm/l	No. dead	Orig. no.
24	5	20	44	8	20	68	9	20	98	2	10	113	8	10		-			-	
18	10	20	23	10	20	25	16	20	39	9	40	68	15	20	66	17	20	109	19	20
25	18	30	40	17	30	63	30	30	101	30	30		-			-			-	
35	11	12	73	10	12	117	12	12		-			-			-			-	
102	22	52	113	10	20		-			-			-			-			-	
38	15	21		-			-			-			-			-			-	
102	57	57	113	19	19		-			-			-			-			-	
113	7	7		-			-			-			-			-			-	
35	0	15	73	0	15	117	12	15		-			-			-			-	
35	17	17	59	15	15	89	16	16		-			-			-			-	
	-			-			-			-			-			-			-	

Part 2 of 2 parts

TABLE III

Summary of the kaolin time-concentration mortality curves presented in Figures 6-13, showing species, estimated 200 hour LC_x in grams/liter of suspended kaolin, the equations from which the estimates were derived, and the coefficient of determination (r^2) of those equations.

Species	200 hour LC_x in gm/l	Equation	Coefficient of determination r^2
Mytilus californianus	LC_{10} = 26	$\ell nY = 22.3 - 3.59\ \ell nX$	0.93
	LC_{20} = 42	$Y = 112 - 0.349X$	0.89
	LC_{50} = 96	$1/Y = 0.0200 + 1.93\ (1/X)$	0.75
Ascidia ceratodes	LC_{10} = 1	$\ell nY = 3.88 - 0.0192X$	0.88
	LC_{20} = 2	$\ell nY = 4.61 - 0.0203X$	0.94
	LC_{50} = 5	$\ell nY = 5.71 - 0.0207X$	0.94
Crangon nigromaculata	LC_{10} = 16	$\ell nY = 5.01 - 0.0113X$	0.76
	LC_{20} = 28	$\ell nY = 5.04 - 0.00850X$	0.87
	LC_{50} = 50	$\ell nY = 7.96 - 0.765\ \ell nX$	0.98
Palaemon macrodactylus	LC_{10} = 24	$\ell nY = 10.3 - 1.34\ \ell nX$	0.71
	LC_{20} = 77	$\ell nY = 4.94 - 0.00300X$	0.96
	LC_{50} = 50% mortality was not reached		
Cancer magister	LC_{10} = 10	$\ell nY = 6.37 - 0.766\ \ell nX$	0.72
	LC_{20} = 18	$\ell nY = 7.01 - 0.776\ \ell nX$	0.96
	LC_{50} = 32	$\ell nY = 3.05 + 83.1\ (1/X)$	0.99
Anisogammarus confervicolus	LC_{10} = 17	$\ell nY = 9.02 - 1.17\ \ell nX$	0.90
	LC_{20} = 35	$1/Y = 0.0377 - 1.79\ (1/X)$	0.97
	LC_{50} = 55	$\ell nY = 3.68 + 67.7\ (1/X)$	0.73
Neanthes succinea	LC_{10} = 9	$Y = 58.6 - 0.246X$	0.88
	LC_{20} = 22	$\ell nY = 4.51 - 0.00700X$	0.92
	LC_{50} = 48	$\ell nY = 5.91 - 0.386\ \ell nX$	0.91
Cymatogaster aggregata (specimens from Bodega Bay)	LC_{10} = 0	$Y = 40.6 - 2.46X$	0.96
	LC_{20} = 0	$Y = 49.3 - 2.93X$	0.98
	LC_{50} = 0	$\ell nY = 4.63 - 0.139X$	0.99
Cymatogaster aggregata (specimens from San Francisco Bay)	LC_{10} = 1	$\ell nY = -0.523 + 154\ (1/X)$	0.91
	LC_{20} = 1	$Y = 6.34 - 0.0270X$	0.82
	LC_{50} = 3	$\ell nY = 0.398 + 138\ (1/X)$	0.79

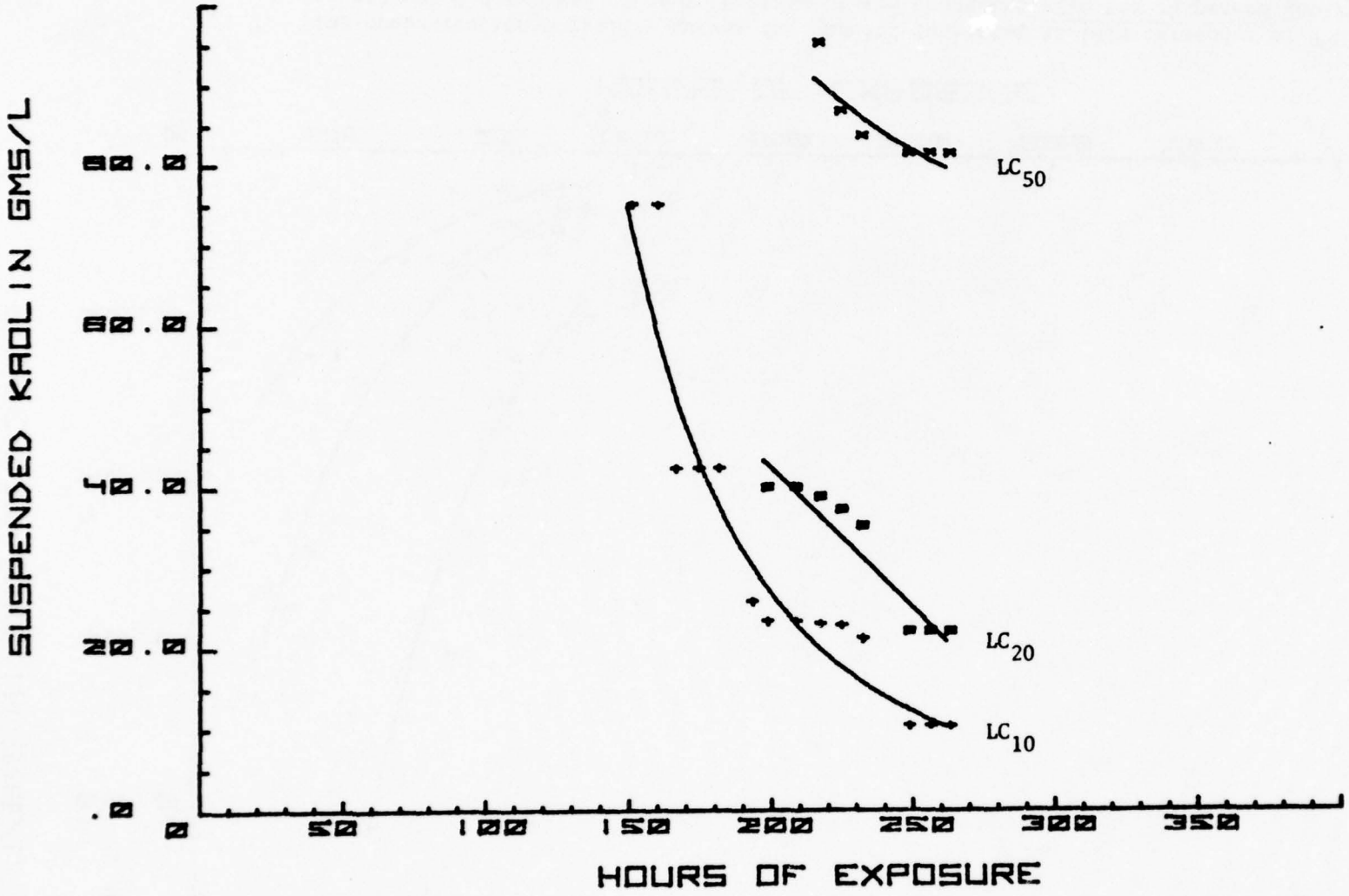


Fig. 6. Time-concentration mortality curves for 10 cm mussels Mytilus californianus at 12°C and saturated dissolved oxygen. Experiment was conducted with six suspended kaolin concentrations from 10 gm/1 to 100 gm/1.

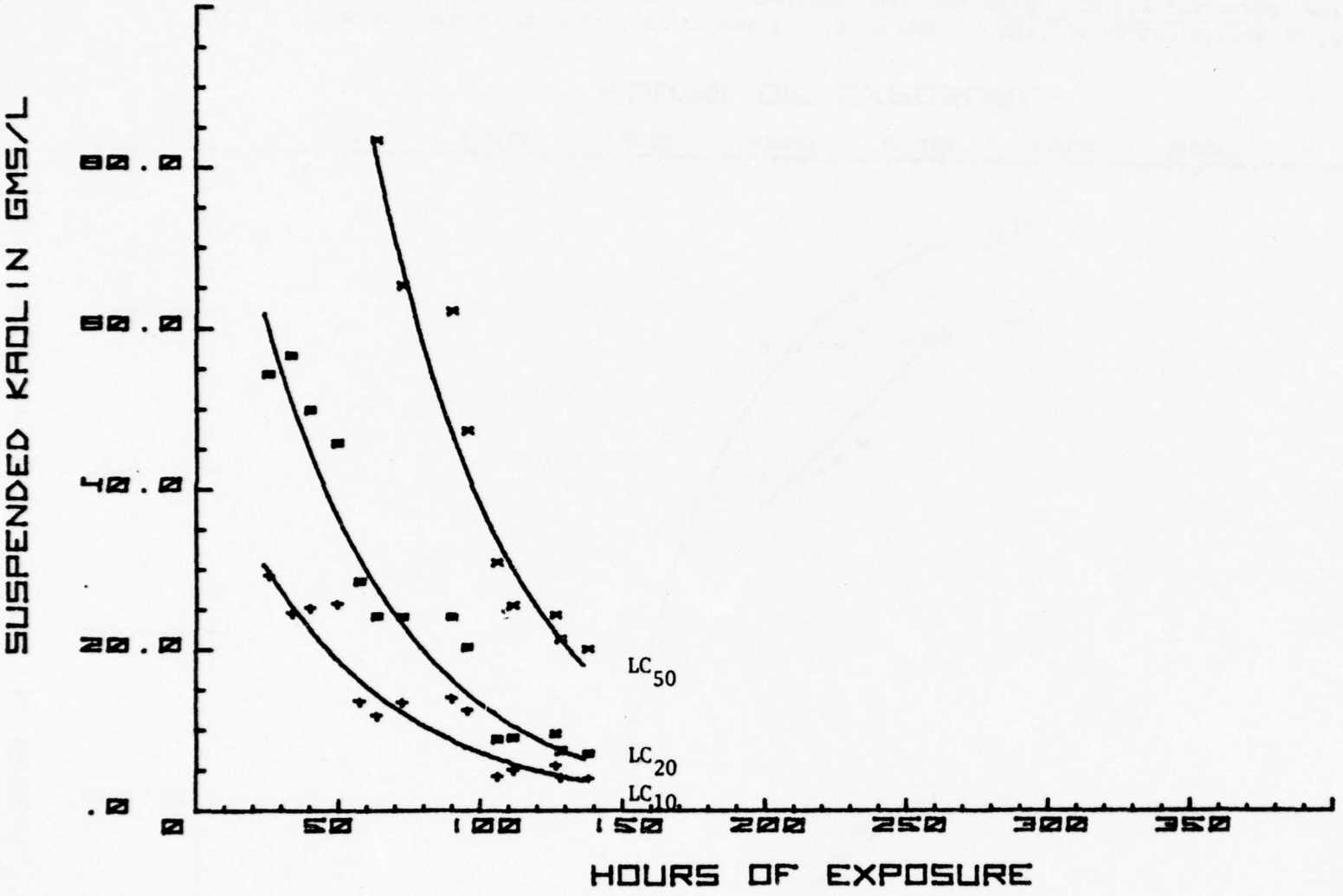


Fig. 7. Time-concentration mortality curves for "adult" tunicates Ascidia ceratodes at 9°C and saturated dissolved oxygen. Experiment was conducted with six suspended kaolin concentrations from 10 gm/1 to 100 gm/1.

response was very different from the two tunicates from San Francisco Bay which had a 288-hour LC_{10} of roughly 100 gm/l (Table I).

The experiment with spot-tailed sand shrimp Crangon nigromaculata (Fig. 8) was continued for 400 hours or over 16 days. Even after this time the mortality curves continued to drop, indicating that a steady state had not been reached, and that with longer exposures the same percentages of animals would have been killed by lower concentrations of suspended kaolin. The 200-hour LC_{50} estimate was 50 gm/l. An experiment was also conducted with C. nigricauda, a related species abundant in San Francisco Bay. This test was characterized by a high incidence of cannibalism of molting shrimp, resulting in high and erratic mortalities (Table II) and making determination of deaths due to the kaolin difficult. The cannibalized molters were therefore dropped from the data before analysis, resulting in low adjusted original numbers and poor accuracy in the LC_x calculations. Even though the final data were too variable to warrant graphical presentation, they did indicate a sensitivity for C. nigricauda somewhere between that for C. franciscorum (Table I) and C. nigromaculata.

The euryhaline shrimp Palaemon macrodactylus (Fig. 9) was less sensitive to suspended kaolin than C. nigromaculata and C. nigricauda. This species was tested for 250 hours and 50% mortality was not reached. The calculated 200-hour LC_{20} for P. macrodactylus was 77 gm/l, higher than the 200-hour LC_{50} for C. nigromaculata.

The other decapod crustacean tested with kaolin was the commercial crab Cancer magister. These crabs, about 5 cm carapace width, were tested for ten days and were more sensitive than any of the

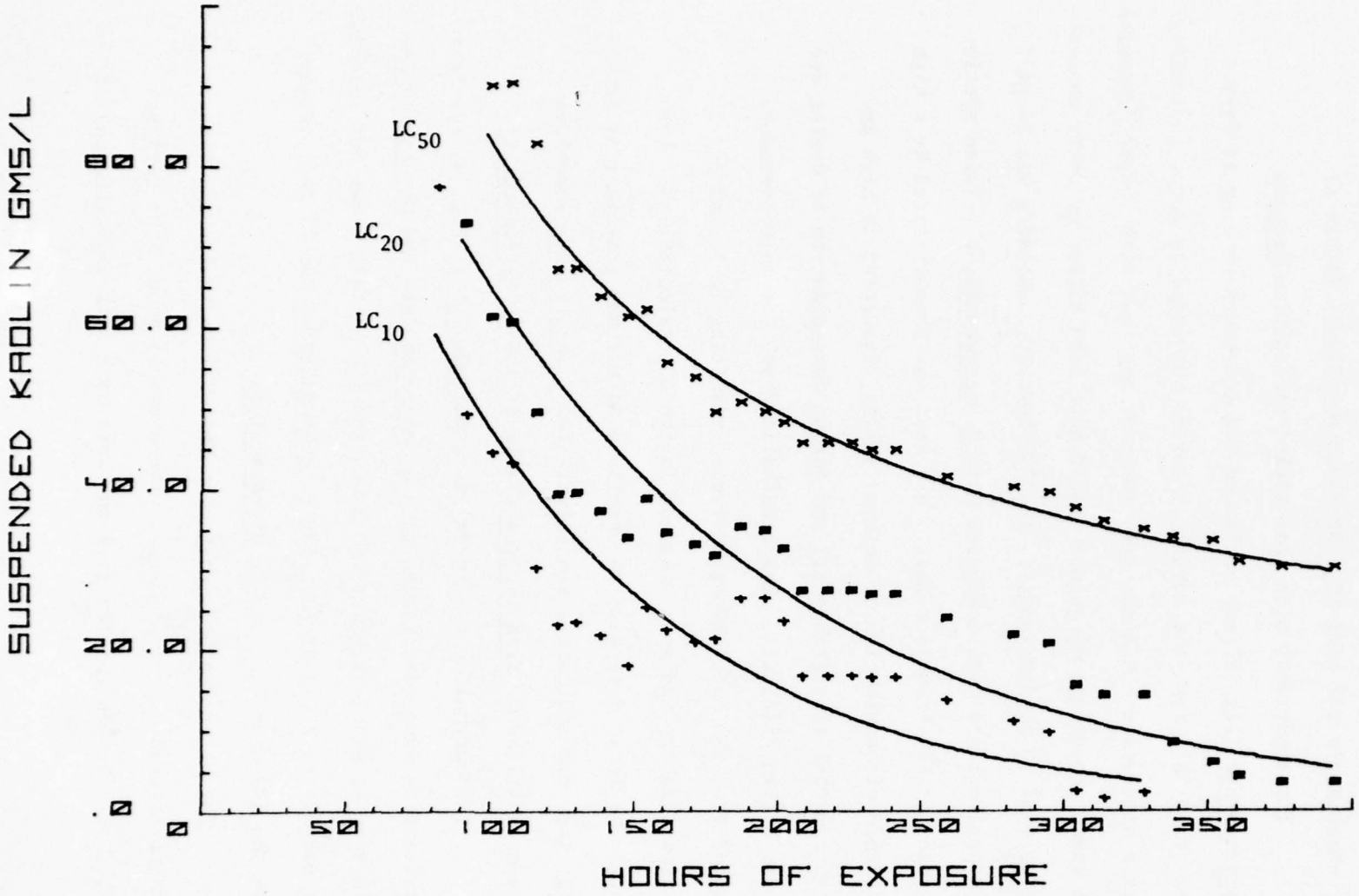


Fig. 8. Time-concentration mortality curves for 6-8.5 cm sand shrimp Crangon nigromaculata at 10°C and saturated dissolved oxygen. Experiment was conducted with six suspended kaolin concentrations from 10 gm/1 to 100 gm/1.

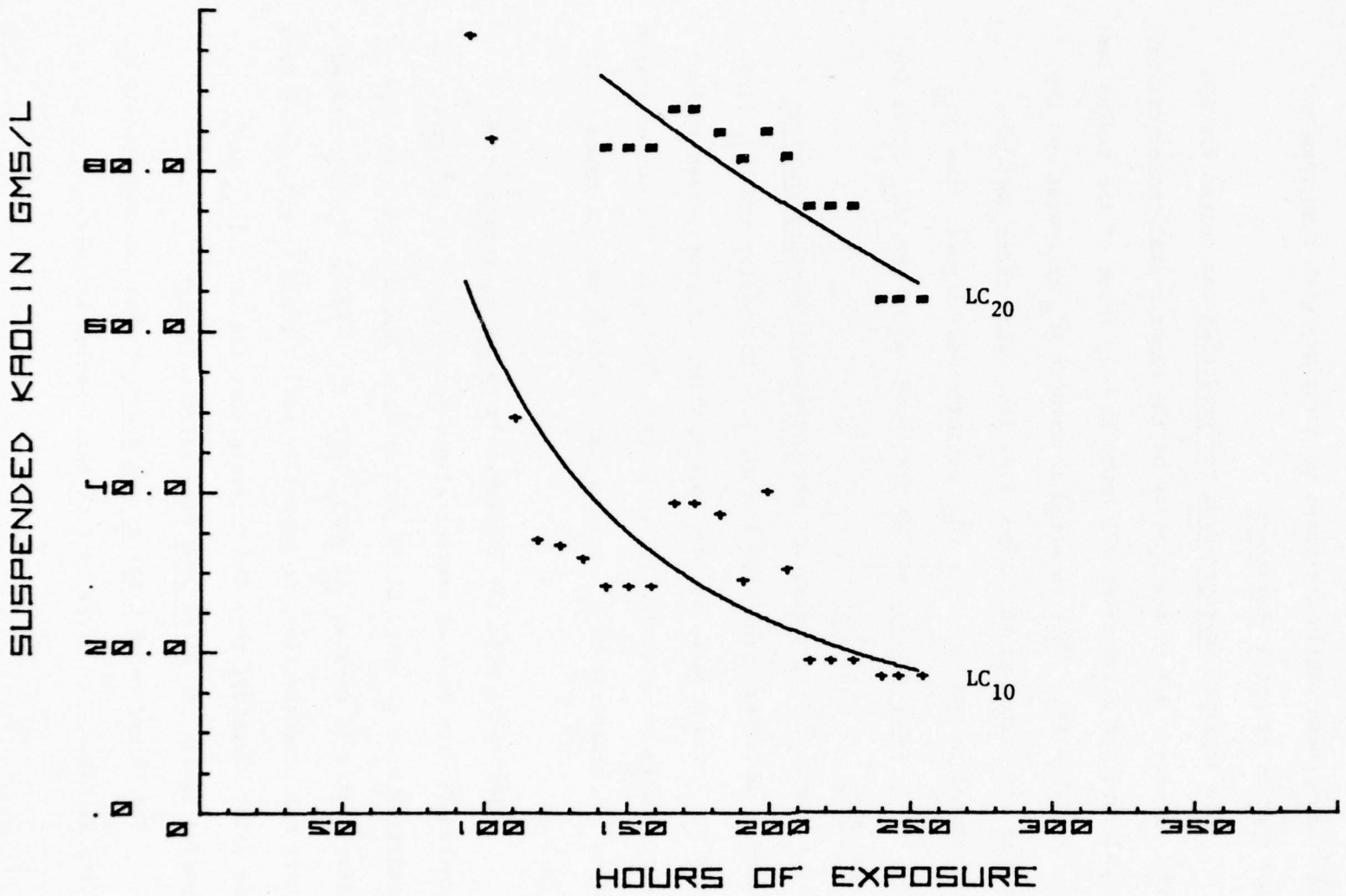


Fig. 9. Time-concentration mortality curves for 2-4 cm shrimp Palaemon macrodactylus at 11°C and saturated dissolved oxygen. Experiment was conducted with six suspended kaolin concentrations from 10 gm/l to 100 gm/l.

shrimp species with a 200-hour LC_{50} of 32 gm/l (Fig. 10). Their tolerance to suspended kaolin decreased more rapidly with time than any other species except A. ceratodes.

The amphipod Anisogammarus confervicolus was tested for 260 hours. However, due to the spacing of the experimental concentrations, animals remained alive after 175 hours in only three of the twelve test aquaria (Table II). This resulted in erratic LC_x estimates and the analysis was halted at that time (Fig. 11). When these data were extrapolated to 200 hours the LC_{50} estimate was 55 gm/l. The LC_{20} curve for A. confervicolus was nearly identical to the LC_{50} curve for C. magister.

The mortality curves for the polychaete Neanthes succinea in suspended kaolin are presented in Fig. 12. In reality the LC_{10} line should not cross 0 gm/l, but due to the choice of test concentrations we were unable to determine its shape below 10 gm/l. The concentration causing 50% mortality of N. succinea in 200 hours was estimated to be 48 gm/l.

Experiments with the English sole Parophrys vetulus were conducted for ten days at concentrations from 10 gm/l to 117 gm/l. No mortalities were observed at 70 gm/l or less, but 80% mortality had occurred after 10 days at 117 gm/l (Table II). Since deaths occurred in only one concentration, no mortality curves could be calculated from these data. However, they did indicate that the LC_{50}, LC_{20} and LC_{10} of suspended kaolin for P. vetulus were relatively high.

The shiner perch Cymatogaster aggregata was the most sensitive species studied. The first test of this species was with organisms

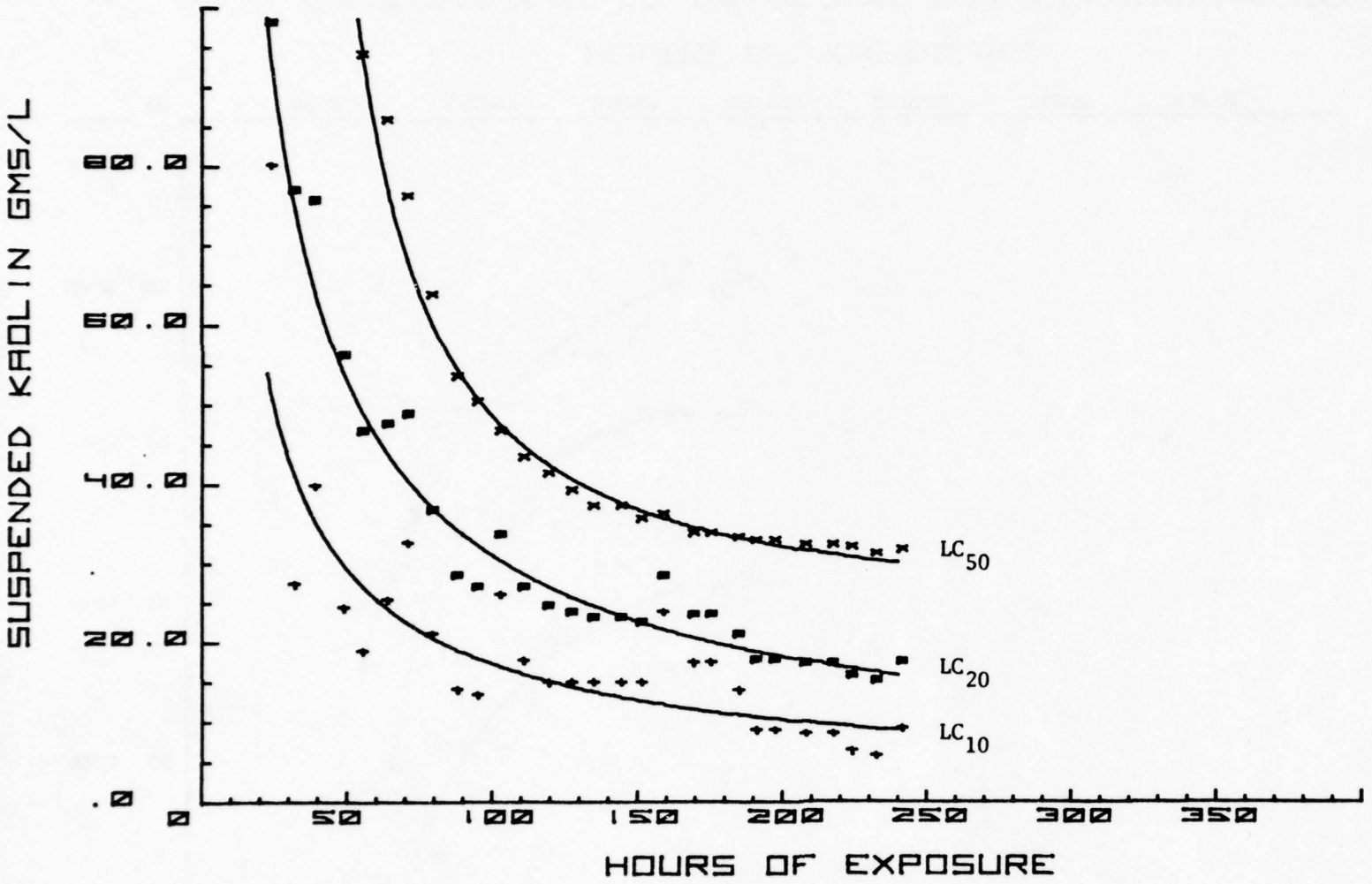


Fig. 10. Time-concentration mortality curves for 5 cm crabs Cancer magister at 10°C and saturated dissolved oxygen. Experiment was conducted with six suspended kaolin concentrations from 10 gm/l to 100 gm/l.

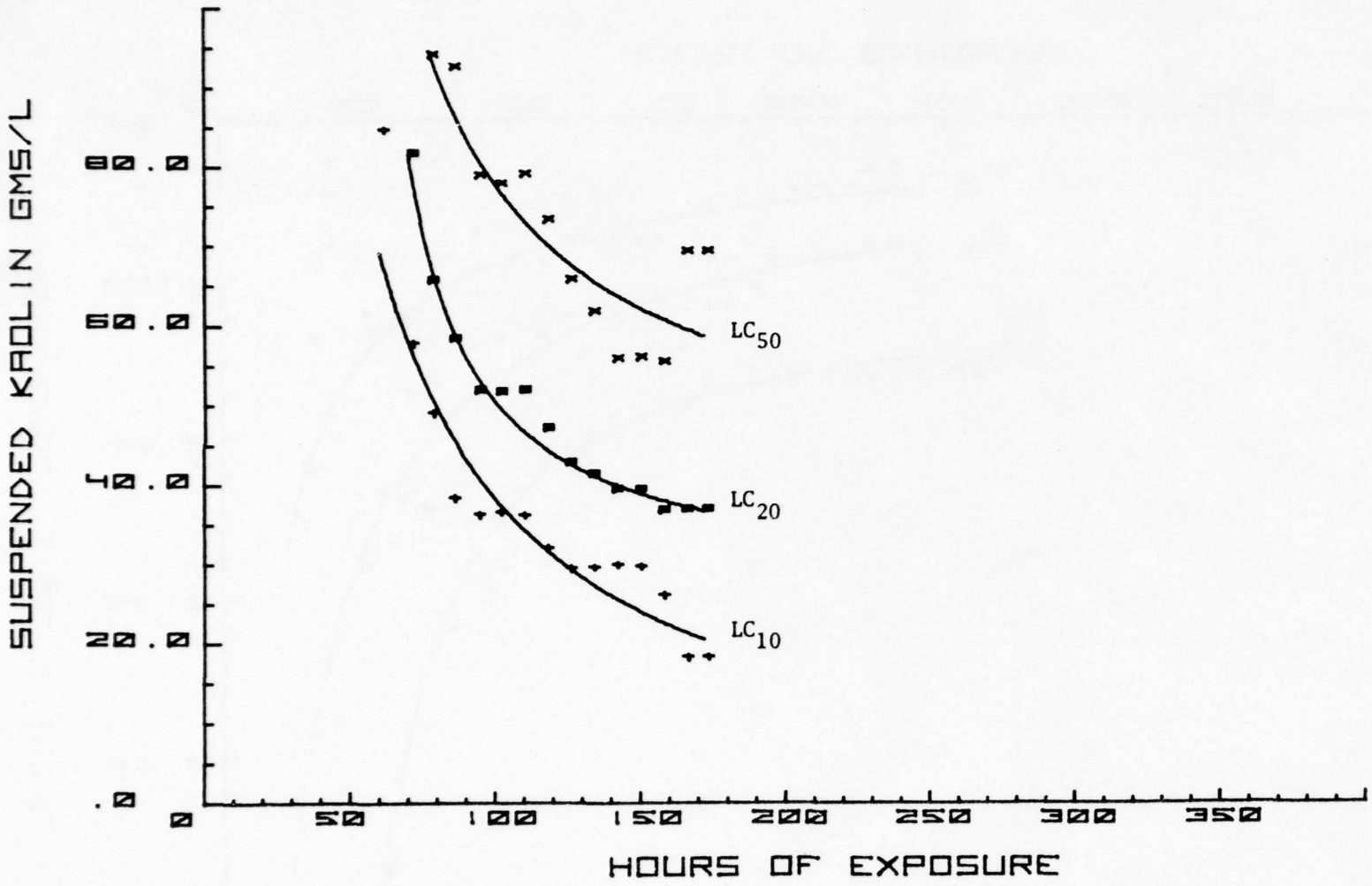


Fig. 11. Time-concentration mortality curves for "adult" amphipods Anisogammarus confervicolus at 11°C and saturated dissolved oxygen. Experiment was conducted with six suspended kaolin concentrations from 10 gm/1 to 100 gm/1.

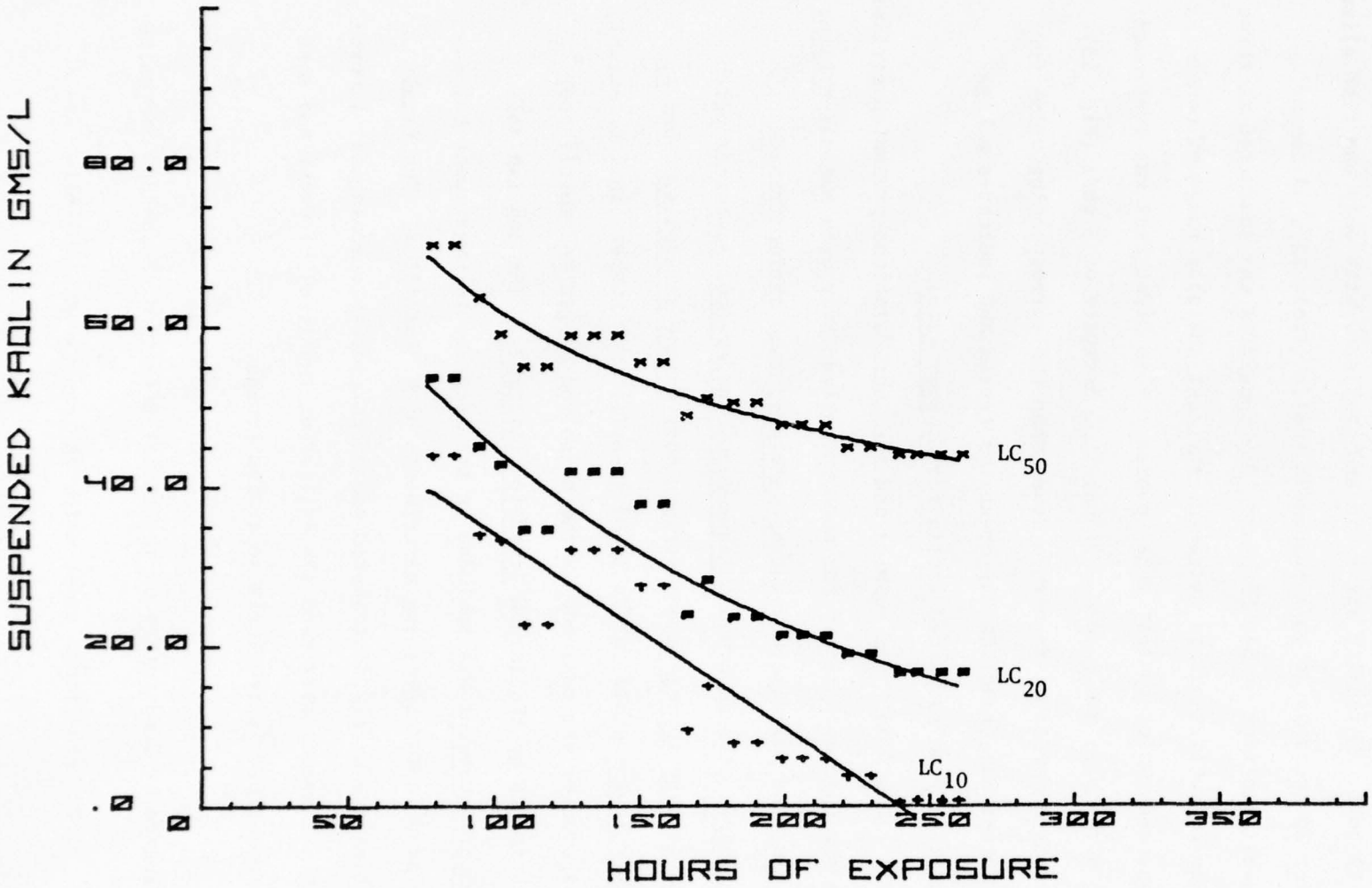


Fig. 12. Time-concentration mortality curves for "adult" polychaetes Neanthes succinea at 11°C and saturated dissolved oxygen. Experiment was conducted with six suspended kaolin concentrations from 10 gm/l to 100 gm/l.

collected at Bodega Bay and tested at a concentration range of 14 gm/l to 89 gm/l. Mortality was rapid and complete, with only one fish alive after 26 hours in 14 gm/l suspended kaolin (Table II). A second abbreviated test of San Francisco Bay specimens was conducted at three lower concentrations to determine the most suitable range of concentrations over which to test this species. This pilot test was continued for eight days and gave a 200-hour LC_{50} estimate of 3 gm/l (Fig. 13). This is an order of magnitude lower than the corresponding value for any other San Francisco Bay organisms tested and indicates a high sensitivity to suspended kaolin for C. aggregata.

Based on these results and the considerations presented earlier, the six species selected for more intensive study were mussels Mytilus edulis, the polychaete Neanthes succinea, sand shrimp Crangon nigricauda, the amphipod Anisogammarus confervicolus, shiner perch Cymatogaster aggregata and striped bass Morone saxatilis. When A. confervicolus could not be found in sufficient numbers in late August, this species was replaced in the dissolved oxygen and multifactor experiments by the isopod Synidotea laticauda. The English sole Parophrys vetulus was included in the temperature experiment but was replaced in the remaining experiments by M. saxatilis. The final species are widely distributed and of considerable ecological importance. Taxonomically none are related at the level of Order and most represent even more widely separated groups.

Influence of Temperature on the Lethal Effects of Suspended Bentonite

The conditions under which the experimental animals were

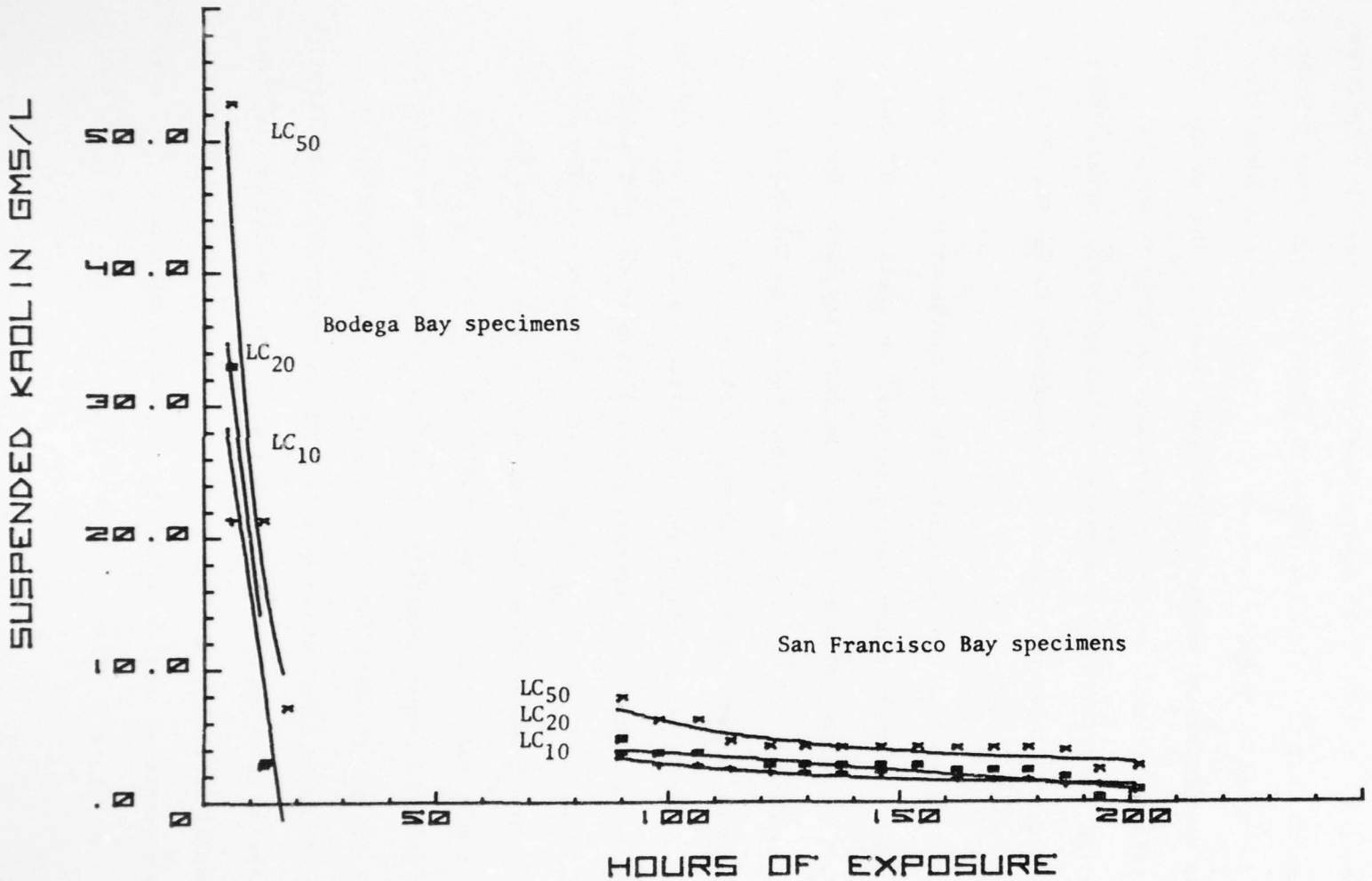


Fig. 13. Time-concentration mortality curves for 8-10 cm shiner perch Cymatogaster aggregata. Specimens from Bodega Bay were tested at 9°C and saturated dissolved oxygen in suspended kaolin concentrations from 10 gm/1 to 100 gm/1. Those from San Francisco Bay were tested at 13°C and saturated dissolved oxygen in suspended kaolin concentrations of 3 gm/1 and 5 gm/1.

collected and held in the laboratory prior to testing are shown in Appendix B, Table BI. The experimental conditions for the temperature tests, conducted at 10°C and 18°C with dissolved oxygen near saturation, are presented in Appendix B, Table BII. This table also identifies those species tested together in the same aquaria. The raw data from the temperature experiments are summarized in Table IV and the equations describing the time-concentration mortality curves, their coefficients of determination and the estimated 10-day LC_x values appear in Table V.

The English sole P. vetulus had no deaths at 10°C in any suspended bentonite concentration and only one death at 18°C during the ten-day experiment (Table IV). Thus no mortality curves could be calculated for this species, which was replaced in the remaining tests by hatchery-reared fingerling striped bass.

The 2-3 cm M. edulis had insignificant deaths in the control tanks and progressively increasing mortalities in the test aquaria at 18°C (Table IV). The LC_{50}, LC_{20} and LC_{10} time-concentration mortality curves for these organisms are presented in Fig. 14. The LC_{10} curve was nearly level after 10 days, while the LC_{20} and LC_{50} curves continued to drop, indicating that longer exposure times might have given lower estimates for these parameters. At 10°C there were significantly fewer deaths than at 18°C, with only two mortalities (8%) occurring at 56 gm/l after 10 days (Table IV). Although no 10° curves could be plotted in Fig. 14, there was clearly a great difference in the response of small M. edulis to suspended bentonite at 10°C and 18°C.

Figure 15 presents plots of the percent survival of M. edulis

TABLE IV

Summary of raw data from mortality tests with suspended bentonite at 18°C and 10°C, showing species, approximate size of animals, test temperature, length of exposure to test conditions and suspended bentonite concentration, total number of deaths and original number of animals in each experimental condition.

Species	Temp. °C	Hours of exposure	Suspended Bentonite Concentrations 1 gm/l	1 No. dead	1 Orig. no.	2 gm/l	2 No. dead	2 Orig. no.	3 gm/l	3 No. dead	3 Orig. no.	4 gm/l	4 No. dead	4 Orig. no.
Parophrys vetulus 7-8 cm	18	236	0	0	32	8	0	16	11	0	16	16	0	16
	10	236	0	0	32	7	0	16	8	0	16	14	0	16
Mytilus edulis 2-3 cm	18	236	0	1	50	8	2	25	11	2	25	16	7	25
	10	236	0	0	50	7	0	25	8	0	25	14	0	25
Crangon nigricauda 2-4.5 cm	18	236	0	0	32	8	1	16	11	0	16	16	2	16
	10	236	0	2	32	7	1	16	8	1	16	14	3	16
Cymatogaster aggregata 3-6 cm	18	195	0	14	32	0.4	12	16	0.6	15	16	1.0	16	16
	10	181	0	16	38	0.4	18	19	0.7	14	19	1.0	18	19
Neanthes succinea "adult"	18	237	0	9	20	7	5	10	8	7	10	15	5	10
	10	237	0	3	20	8	4	10	10	0	10	15	5	10
Ani_ogammarus confervicolus "adult"	18	237	0	4	6	7	1	9	8	4	8	15	2	8
	10	237	0	1	24	8	4	12	10	2	13	15	6	14

Table IV, continued

Species	Temp. °C	Suspended Bentonite Concentrations 5 gm/l	5 No. dead	5 Orig. no.	6 gm/l	6 No. dead	6 Orig. no.	7 gm/l	7 No. dead	7 Orig. no.
Parophrys vetulus	18	25	1	16	36	0	16	54	0	16
	10	22	0	16	36	0	16	56	0	16
Mytilus edulis	18	25	16	25	36	15	25	54	18	20
	10	22	0	25	36	0	25	56	2	25
Crangon nigricauda	18	25	0	16	36	4	16	54	1	16
	10	22	1	16	36	1	16	56	1	16
Cymatogaster aggregata	18	1.6	16	16	3.2	15	16	3.8	16	16
	10	1.5	17	19	3.3	19	19	4.2	19	19
Neanthes succinea	18	22	4	10	36	5	10	48	7	10
	10	22	3	10	31	0	10	50	2	10
Anisogammarus confervicolus	18	22	5	11	36	3	19	48	7	12
	10	22	7	8	31	6	10	50	4	13

TABLE V

Summary of the suspended bentonite-temperature mortality curves, showing species, temperature, estimated 10-day (240 hour) LC_x in grams/liter of suspended bentonite, the equations from which the estimates were derived, and the coefficient of determination (r^2) of those equations.

Species	Temp.	10-day LC_x in gm/l	Equation	Coefficient of determination (r^2)
Mytilus edulis	(18°C)	LC_{10} = 10	1/Y = .1630 - 12.40 (1/X)	0.86
		LC_{20} = 14	lnY = 1.43 + 251 (1/X)	0.91
		LC_{50} = 25	1/Y = 0.0836 - 10.4 (1/X)	0.93
Crangon nigricauda	(18°C)	LC_{10} = 9.4	lnY = 2.750 - .00210 (X)	0.79
		LC_{20} = 57	lnY = 4.07 - 7.01 (1/X)	0.75
		LC_{50}	50% mortality was not reached	

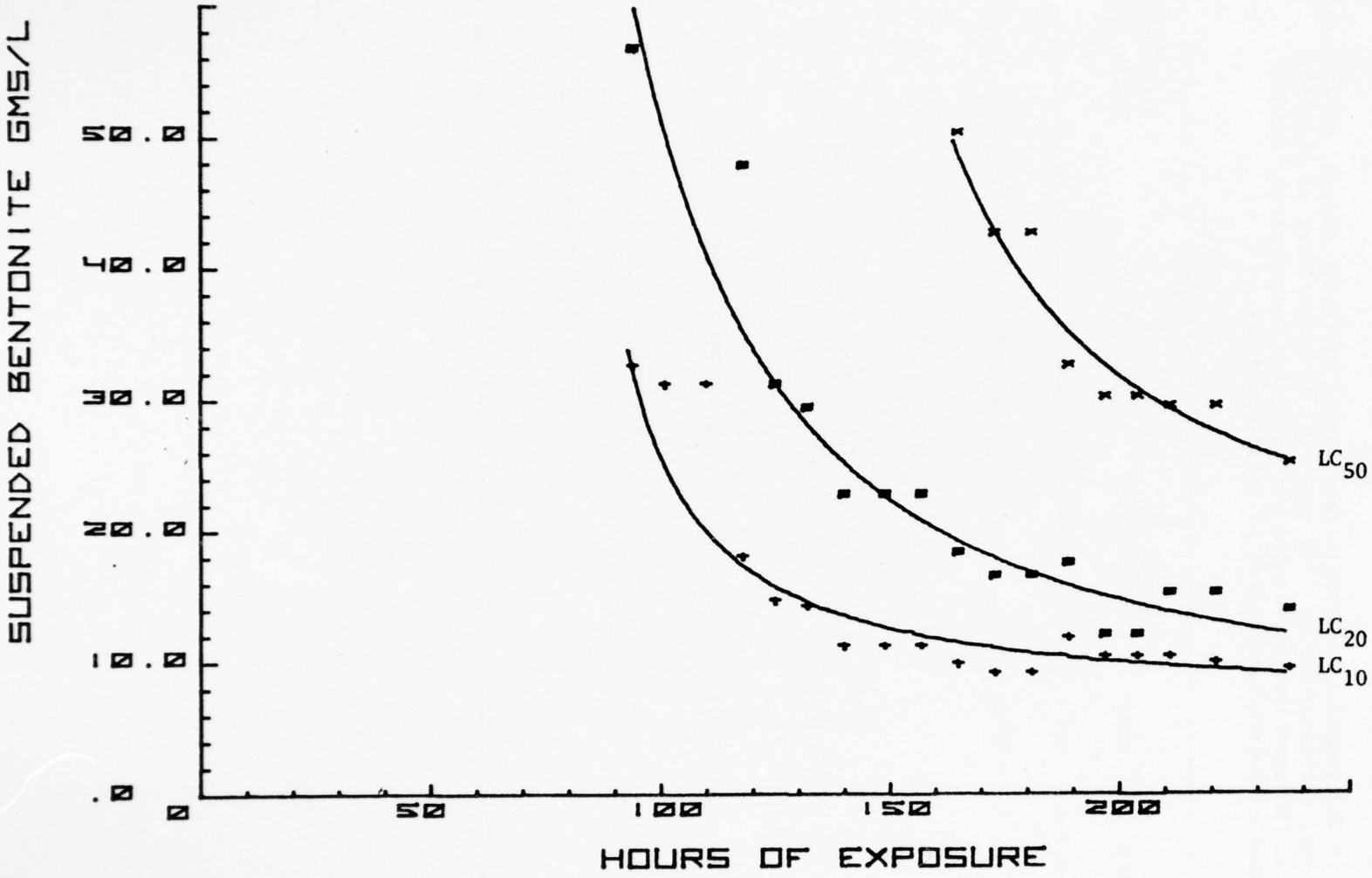


Fig. 14. Time-concentration mortality curves for 2-3 cm long mussels Mytilus edulis at 18°C and saturated dissolved oxygen. Experiment was conducted with six suspended bentonite concentrations from 6 gm/1 to 60 gm/1.

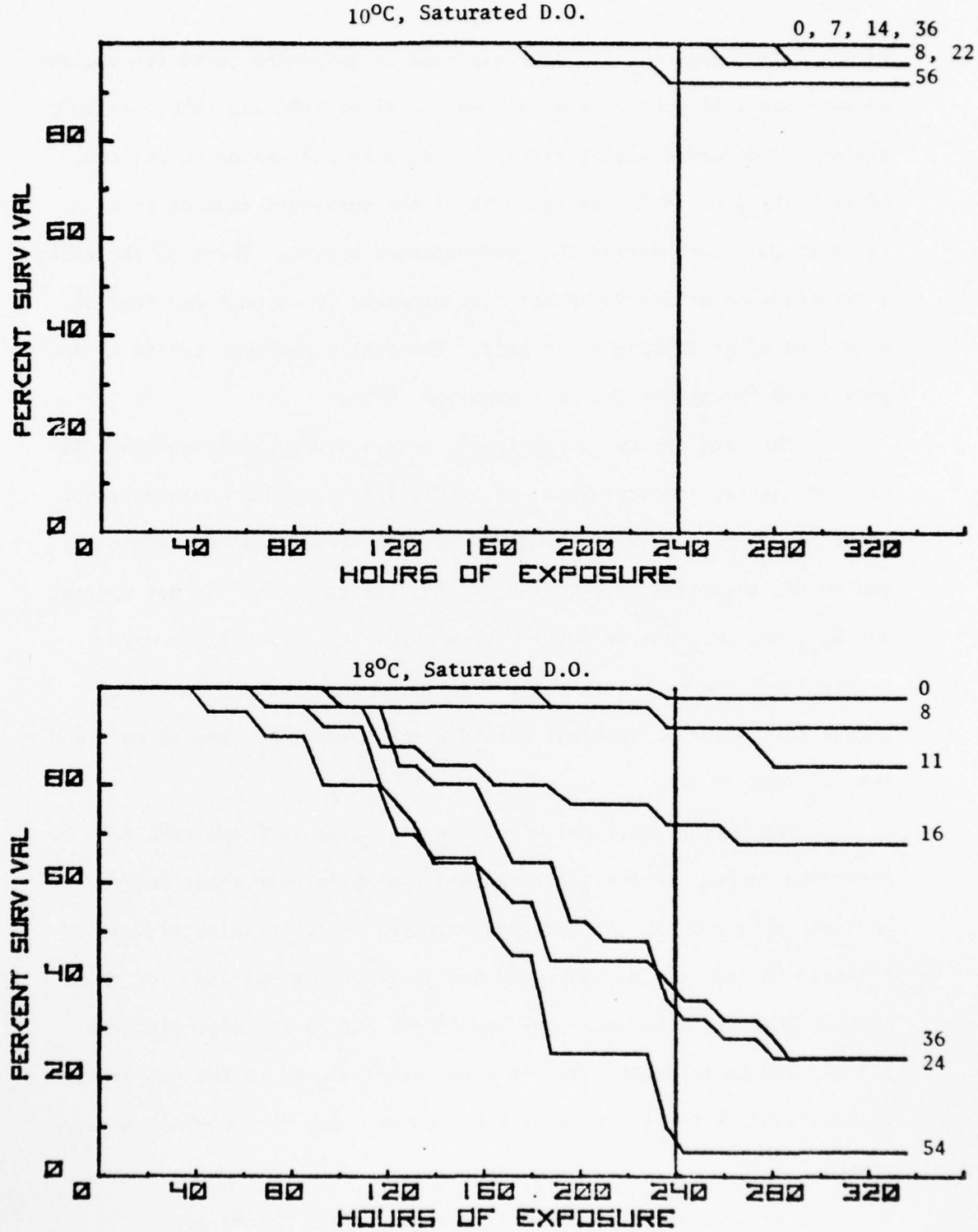


Fig. 15. Percent survival of 2-3 cm Mytilus edulis during 240 hours of exposure to suspended bentonite and an additional 96 hour post-exposure period with the survivors in clear water. Experiments were done at 10°C and 18°C at saturated dissolved oxygen. Numbers on the lines are suspended bentonite concentrations in gm/l.

versus time during the 240-hour exposure to suspended bentonite and for an additional 96-hour period in clear water at 10°C and 18°C. At 10°C few deaths occurred during either the test or post-exposure periods. After testing at 18°C less than 10% of the survivors exposed to 0, 8, 11 or 16 gm/l died during the post-exposure period. Three of the eight survivors died within 96 hours after exposure to 24 gm/l and four of nine died after testing at 36 gm/l. The single survivor tested at 54 gm/l lived throughout the post-exposure period.

The sand shrimp C. nigricauda also had very different mortalities at the two temperatures. At 10°C deaths exceeded 6% in only one aquarium, were scattered throughout all the concentrations (Table IV) and no LC_x estimates were made. At 18°C 50% mortality was not reached but LC_{20} and LC_{10} curves were derived (Fig. 16). Both lines were nearly level and much more widely separated than the LC_{10} and LC_{20} curves for any other species. At 18°C the 10-day LC_{20} was 55 gm/l and the LC_{10} was 10 gm/l.

The percent survival of C. nigricauda at 10°C and 18°C is presented in Fig. 17 for the suspended bentonite experiment and the post-exposure period. At both temperatures 12 to 16 animals survived exposure to each of the suspended bentonite concentrations. Of these no more than two died under any conditions during the post-exposure period, and these deaths did not seem to be related to the temperature or suspended bentonite concentrations under which the animals had been tested.

The most sensitive species tested was the shiner perch C. aggregata. The experiments were terminated at approximately 200 hours

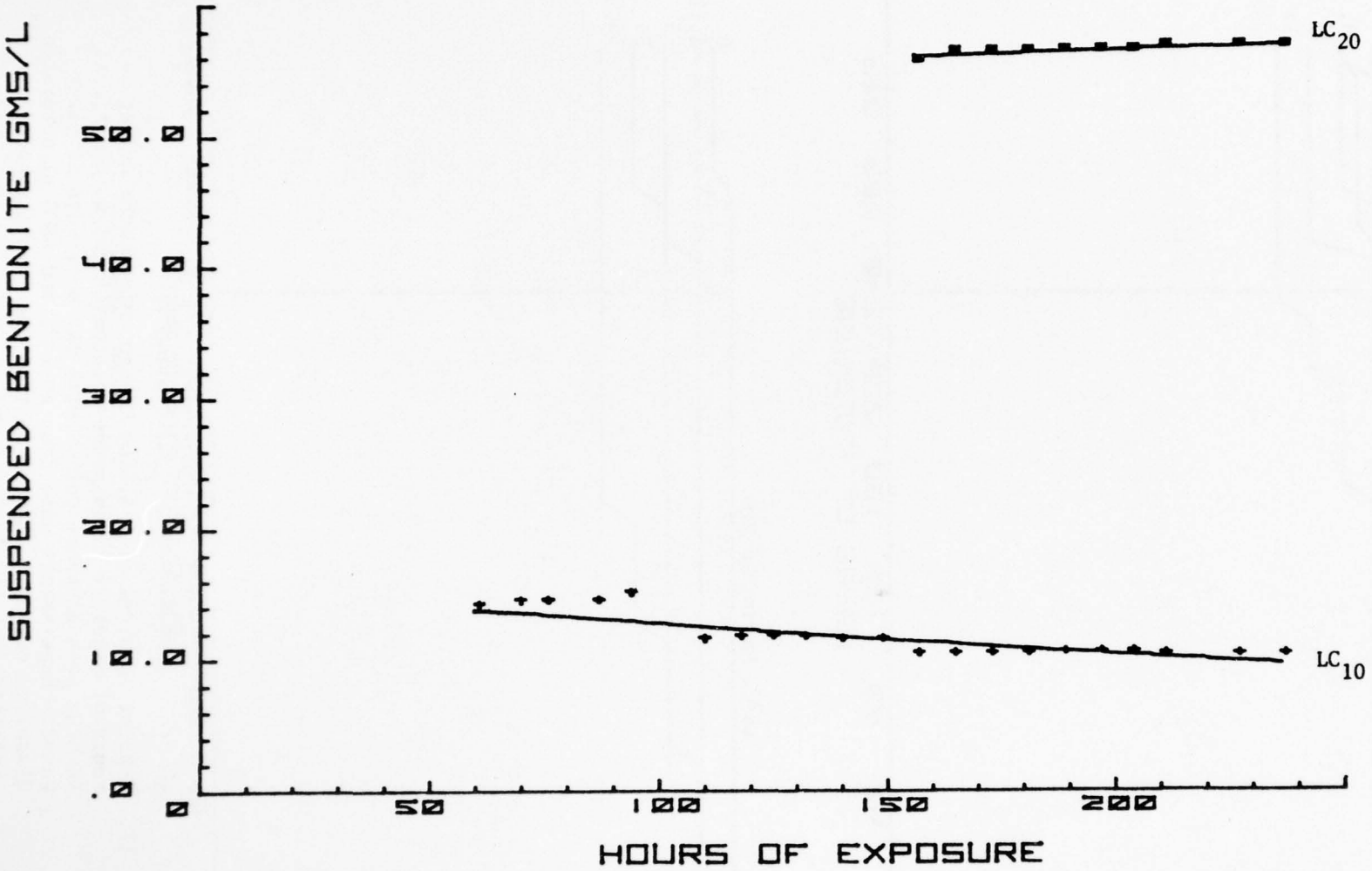


Fig. 16. Time-concentration mortality curves for 2-4.5 cm sand shrimp Crangon nigricauda at 18°C and saturated dissolved oxygen. Experiment was conducted with six suspended bentonite concentrations from 6 gm/1 to 60 gm/1.

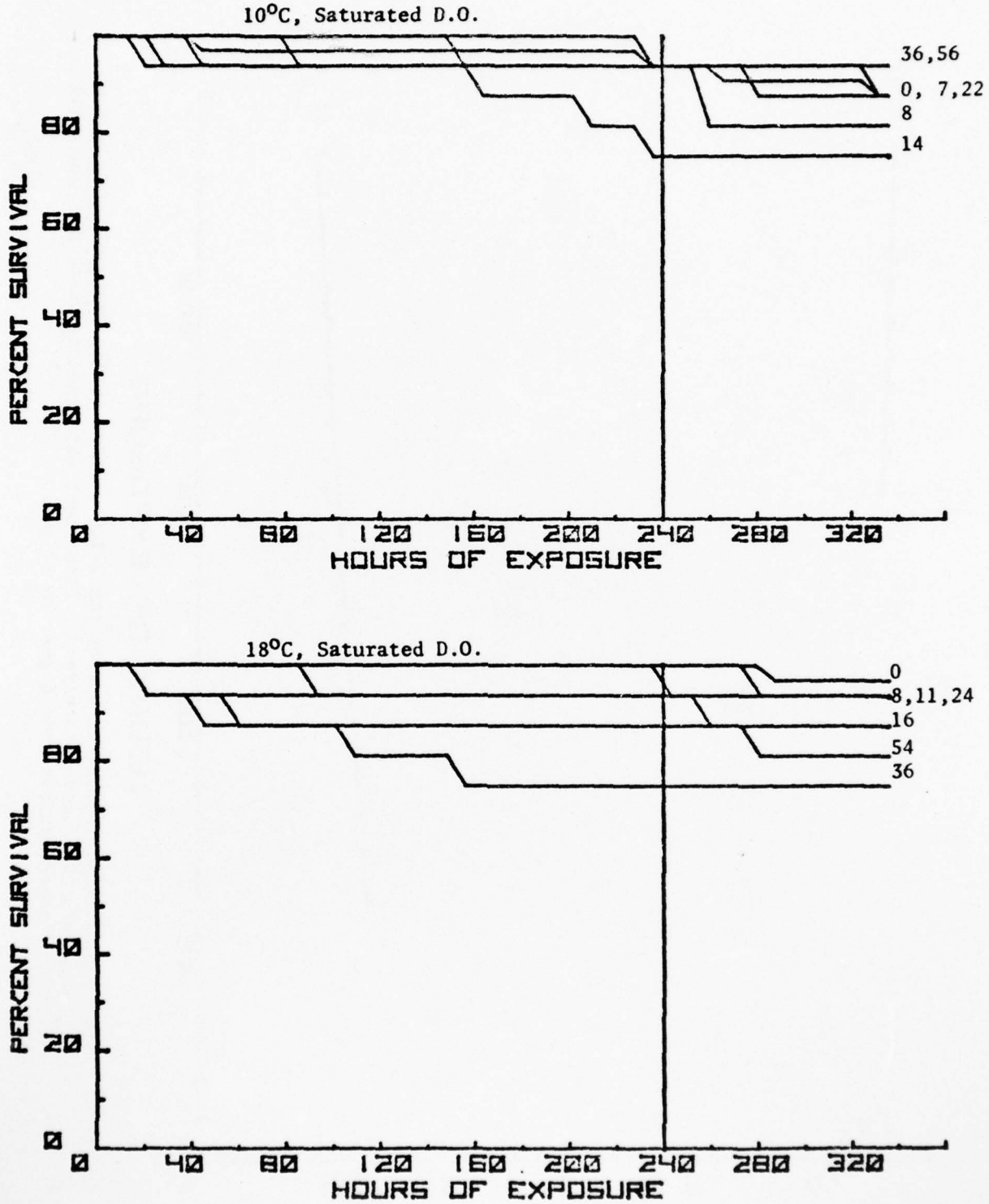


Fig. 17. Percent survival of 2-4.5 cm Crangon nigricauda during 240 hours of exposure to suspended bentonite and an additional 96-hour post-exposure period with the survivors in clear water. Experiments were done at 10°C and 18°C at saturated dissolved oxygen. Numbers on the lines are suspended bentonite concentrations in gm/l.

of exposure time when almost total mortality has occurred in the test aquaria and mortality in the control tanks exceeded 40% (Table IV). Adjustments were made for the control mortalities but these data were clearly biased by the influence of significant stresses other than suspended bentonite. In subsequent experiments the high control mortalities were eliminated, indicating a reduction in uncontrolled stresses, by checking the fish only once per day and feeding brine shrimp daily.

The response of the polychaete N. succinea at both temperatures was too erratic (Table IV) to permit calculation of reliable LC_x estimates and did not seem to be related to the physical parameters monitored in the aquaria (Appendix B, Table BII). The data seemed to indicate more mortalities in the presence of suspended particles than in clear water, but were so variable that a more specific statement was unwarranted.

A large number of amphipods A. confervicolus escaped from the baskets and were lost, requiring considerable adjustments in the data. The observed responses were very erratic and showed little correlation with concentration. Therefore mortality curves for this species are not presented. In subsequent experiments the design of the fine mesh baskets was improved to reduce escapes.

Influence of Dissolved Oxygen on the Lethal Effects of Suspended Bentonite

The conditions under which the experimental animals were collected, held in the laboratory and tested in these experiments,

conducted with 2 ppm and 5 ppm dissolved oxygen at 18°C, are presented in Appendix C. Those species tested in the same aquaria are identified in Table CII. The results of the dissolved oxygen experiments are summarized in Table VI and the equations for the resulting LC_x curves, their coefficients of determination and the estimated 10-day LC_x values are presented in Table VII. In these experiments the control aquaria were maintained at saturated dissolved oxygen, so that the adjusted deaths in the test aquaria, and thus the graphs, represent the combined effects of simultaneously raised suspended solids and lowered dissolved oxygen.

More M. edulis were killed by lower concentrations of suspended bentonite at 2 ppm dissolved oxygen than at 5 ppm (Figs. 18 and 19). Significant deaths were not noted until late in the experiment but then occurred rapidly. The 240-hour estimated LC_x values at 2 ppm dissolved oxygen were only 33% of 50% of the corresponding values at 5 ppm.

The survival of the remaining mussels when transferred to clear, oxygen saturated water at the same temperature is illustrated in Fig. 20. After testing at 2 ppm dissolved oxygen, more than 85% of the survivors from most bentonite concentrations lived at least 96 hours. The exceptions were those which had been exposed to 14 gm/l, where three of nine survivors died, and those exposed to 21 gm/l where four of five survivors died. After the experiment at 5 ppm oxygen, 25% to 40% mortality occurred among mussels exposed to 15 gm/l, 21 gm/l and 35 gm/l, while 66% of the survivors died in 96 hours after exposure to 48 gm/l.

The influence of dissolved oxygen on suspended solids tolerance

Table VI

Summary of raw data from mortality tests with suspended bentonite at 5 ppm and 2 ppm dissolved oxygen, showing species, approximate size of animals, experimental dissolved oxygen levels, length of exposure to test conditions and the suspended solids concentration, total number of deaths and the original number of animals in each experimental condition. Oxygen in the control aquaria was maintained at saturation.

			Suspended Bentonite Concentrations											
			1			2			3			4		
Species	D.O. ppm	Hours of Exp.	gm/l	No. Dead	Orig. No.	gm/l	No. Dead	Orig. No.	gm/l	No. Dead	Orig. No.	gm/l	No. Dead	Orig. No.
Mytilus edulis 2-3 cm	5	239	0	0	32	6	3	31	8	1	32	15	16	32
	2	239	0	1	32	6	10	32	9	18	32	14	20	32
Crangon nigricauda 3-4 cm	5	236	0	0	16	7	6	15		*		15	6	16
	2	236	0	0	16	8	11	16	10	14	16	15	16	16
Synidotea laticauda "adult"	5	236	0	1	20	7	1	19	8	5	18	15	2	20
	2	236	0	1	20	8	3	19	10	3	19		*	
Neanthes succinea "adult"	5	236	0	3	12	7	5	9	8	5	9	15	3	9
	2	236	0	2	16	8	4	9	10	4	9		*	
Cymatogaster aggregata 5.5-7 cm	5	239	0	0	15	0.2	0	15	0.3	0	15	0.5	1	15
	2	236	0	0	15	0.6	1	15	0.8	7	15	1.3	13	15
Morone saxatilis 5-8 cm	5	236	0	0	15	0.2	0	15	0.3	0	15	0.5	0	15
	2	236	0	0	15	0.6	0	15	0.8	1	15	1.3	1	15

Table VI, continued

Species	D.O. ppm	Suspended Bentonite Concentrations 5 gm/l	5 No. Dead	5 Orig. No.	6 gm/l	6 No. Dead	6 Orig. No.	7 gm/l	7 No. Dead	7 Orig. No.
Mytilus edulis	5	21	14	33	35	22	32	48	24	32
	2	21	25	32	30	25	32	46	23	32
Crangon nigricauda	5	25	14	16	35	11	16	47	13	16
	2	19	16	16	33	16	16	43	16	16
Synidotea laticauda	5	25	3	20	36	7	20	47	6	20
	2	21	4	20	33	2	20	49	3	20
Neanthes succinea	5	25	4	9	36	3	9	47	3	9
	2	21	5	9	33	2	9	49	2	9
Cymatogaster aggregata	5	0.6	0	15	1.1	3	15	2.0	3	15
	2		*		4.2	15	15		*	
Morone saxatilis	5	0.6	0	15	1.1	0	15	2.0	0	15
	2		*		4.2	7	15		*	

* These aquaria were dropped from the analyses when their dissolved oxygen controls malfunctioned, rapidly altering the experimental conditions.

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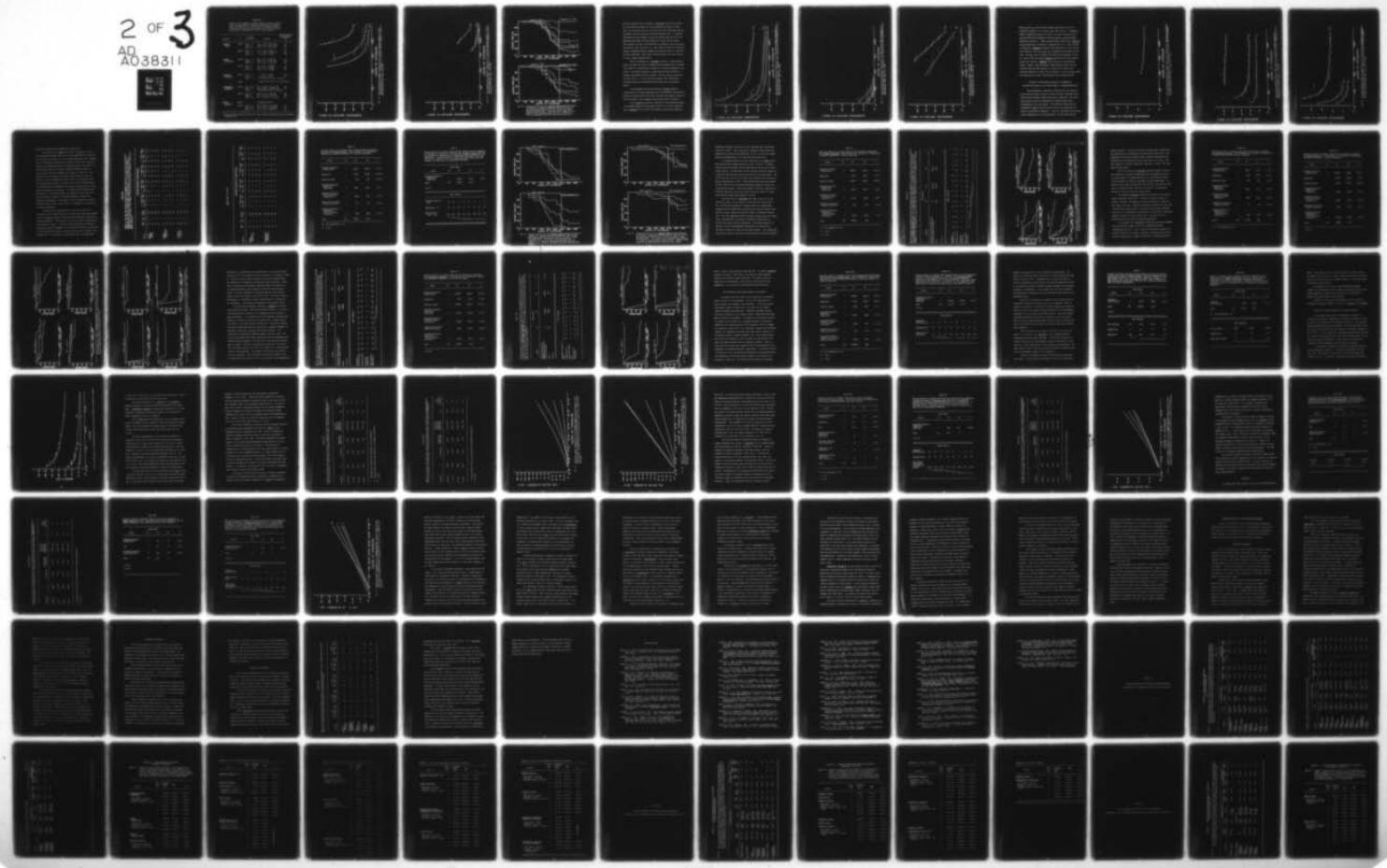

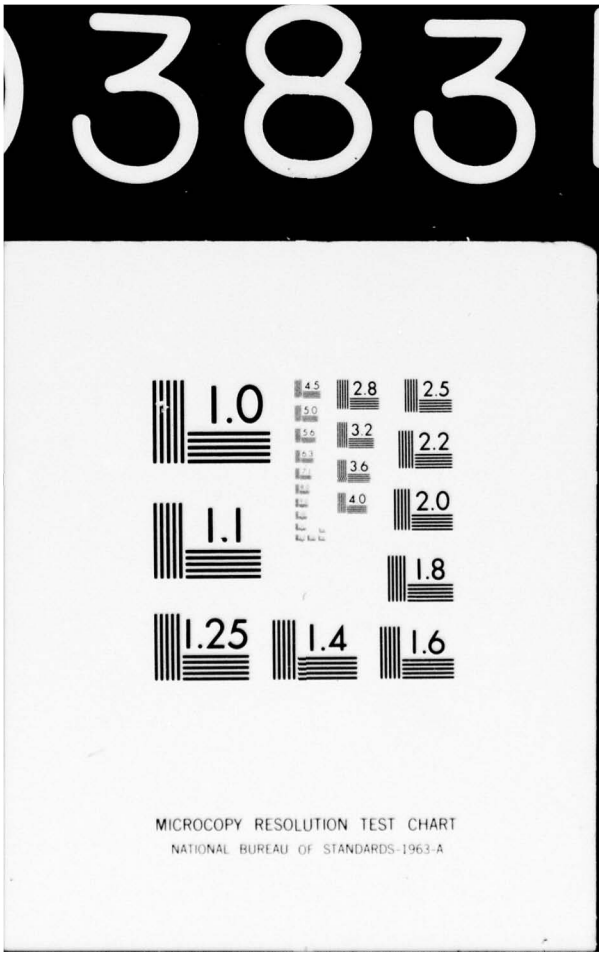
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Table VII

Summary of the suspended bentonite-dissolved oxygen mortality curves, showing species, dissolved oxygen level, estimated 10-day (240 hour) LC_x in grams/liter of suspended bentonite, the equations from which the estimates were derived, and the coefficients of determination (r^2) of those equations.

Species	D.O.	10-day LC_x in gm/l	Equation	coefficient of determination (r^2)
Mytilus edulis	(5ppm)	LC_{10} = 2	$\ell nY = -2.01 + 702\ (1/X)$	.79
		LC_{20} = 6	$\ell nY = -1.07 + 686\ (1/X)$	.83
		LC_{50} = 20	$1/Y = 0.77 - 30.4\ (1/X)$	.92
	(2ppm)	LC_{10} = 1	$Y = 6.05 - 0.0231X$	.69
		LC_{20} = 2	$\ell nY = -2.88 + 838\ (1/X)$	.92
		LC_{50} = 9	$Y = 83.6 - 0.309X$	.98
Crangon nigricauda	(5ppm)	LC_{10} = 1	$\ell nY = 5.01 - 0.854\ \ell nX$	.80
		LC_{20} = 3	$\ell nY = 5.07 - 0.712\ \ell nX$	.84
		LC_{50} = 13	$\ell nY = 5.19 - 0.474\ \ell nX$	.88
	(2ppm)	LC_{10} = *	$\ell nY = 2.83 - 0.0856X$	.83
		LC_{20} = *	$\ell nY = 2.93 - 0.0681X$	.85
		LC_{50} = *	$\ell nY = 3.12 - 0.0384X$	.89
Synidotea laticauda	(5ppm)	LC_{10} = 6	$Y = 63.8 - 0.241X$	.87
		LC_{20} = 16	$Y = 105 - 0.370X$	.98
		LC_{50} =	50% mortality was not reached	
	(2ppm)		Insufficient deaths for calculation	
Cymatogaster aggregata	(5ppm)	LC_{10} = 0.5	$\ell nY = -0.818 + 43.2\ (1/X)$	.80
		LC_{20} = 1.8	$\ell nY = 0.568 - .000309\ \ell nX$	.78
		LC_{50} =	50% mortality was not reached	
	(2ppm)	LC_{10} = *	$\ell nY = 0.704 - 5.30\ \ell nX$	.80
		LC_{20} = *	$\ell nY = 1.42 - 0.557\ \ell nX$	.88
		LC_{50} = 0.9	$\ell nY = -0.238 + 21.3\ (1/X)$	.80
Morone saxatilis	(5ppm)		No deaths occurred	
	(2ppm)	LC_{10} = 1.2	$\ell nY = 0.146 + 7.54\ (1/X)$	.51
		LC_{20} = 2.0	$1/Y = 0.544 - 8.57\ (1/X)$	.87
		LC_{50} = 4.6	$\ell nY = 1.63 - 0.000458X$	.65

* After less than 100 hours most of the test animals had died and the LC_x estimates became erratic.

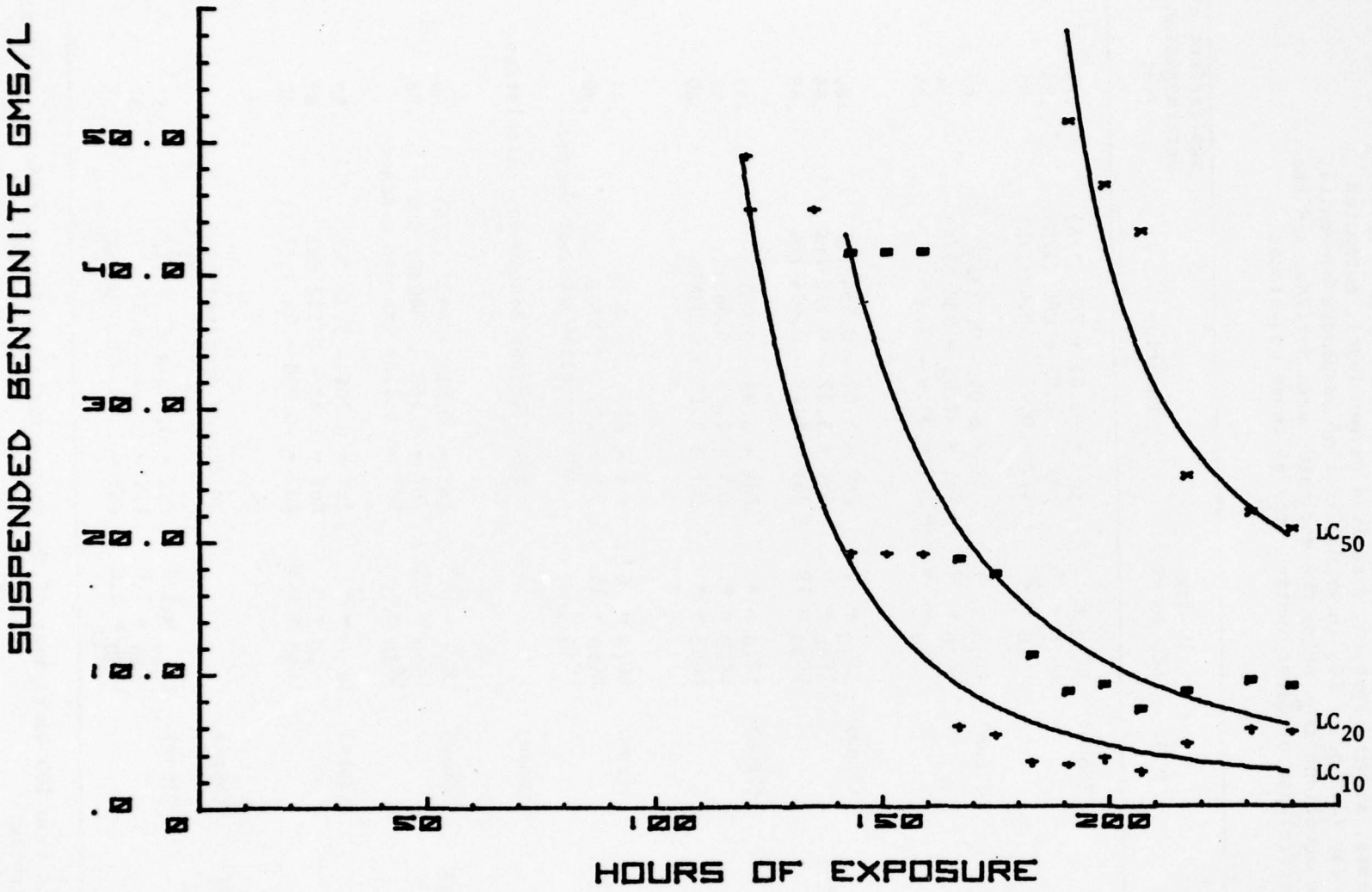


Fig. 18. Time-concentration mortality curves for 2-3 cm mussels Mytilus edulis at 5 ppm dissolved oxygen and 18°C. Experiment was conducted with six suspended bentonite concentrations from 6 gm/1 to 60 gm/1.

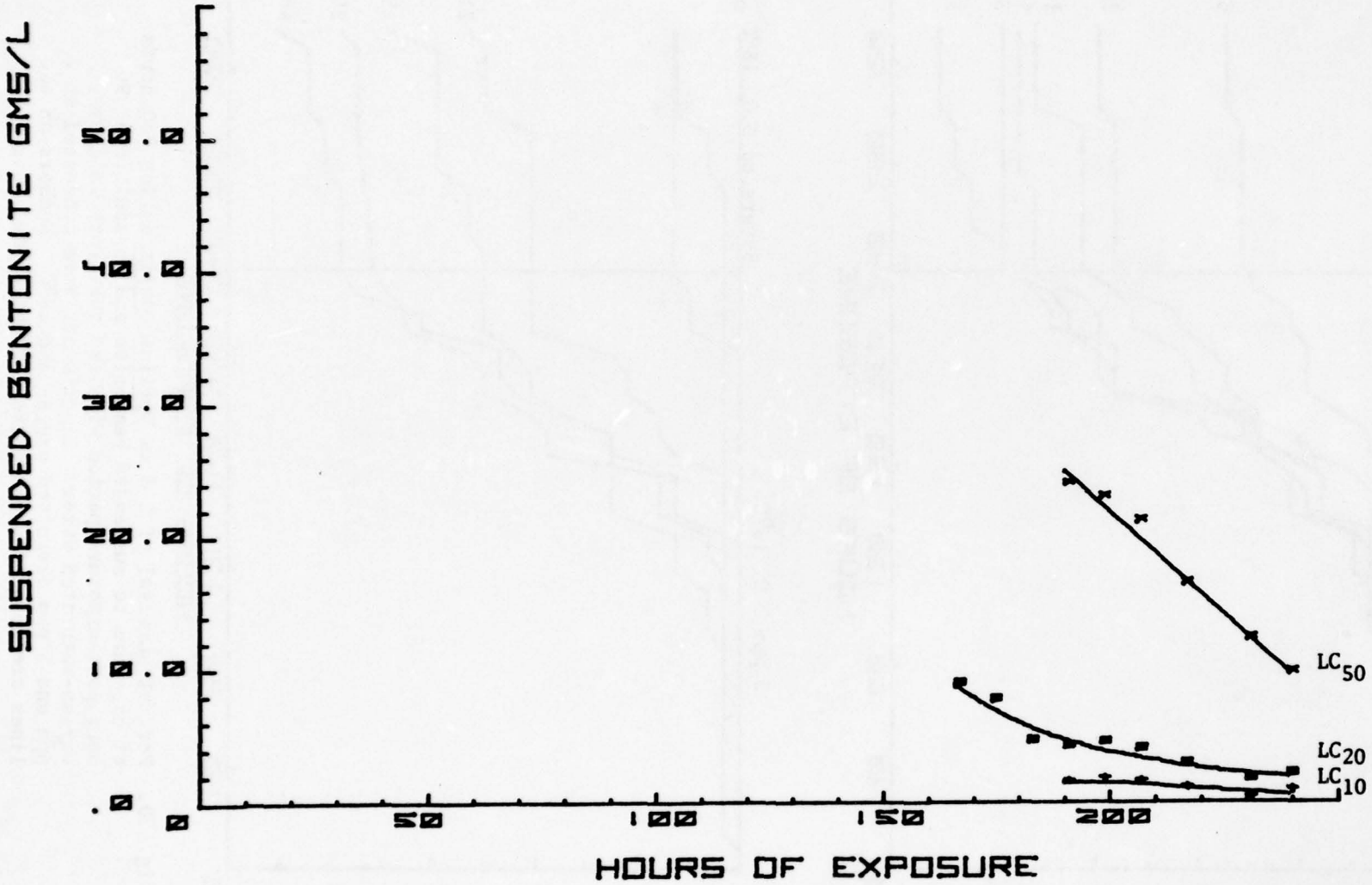


Fig. 19. Time-concentration mortality curves for 2-3 cm mussels Mytilus edulis at 2 ppm dissolved oxygen and 18°C. Experiment was conducted with six suspended bentonite concentrations from 6 gm/1 to 60 gm/1.

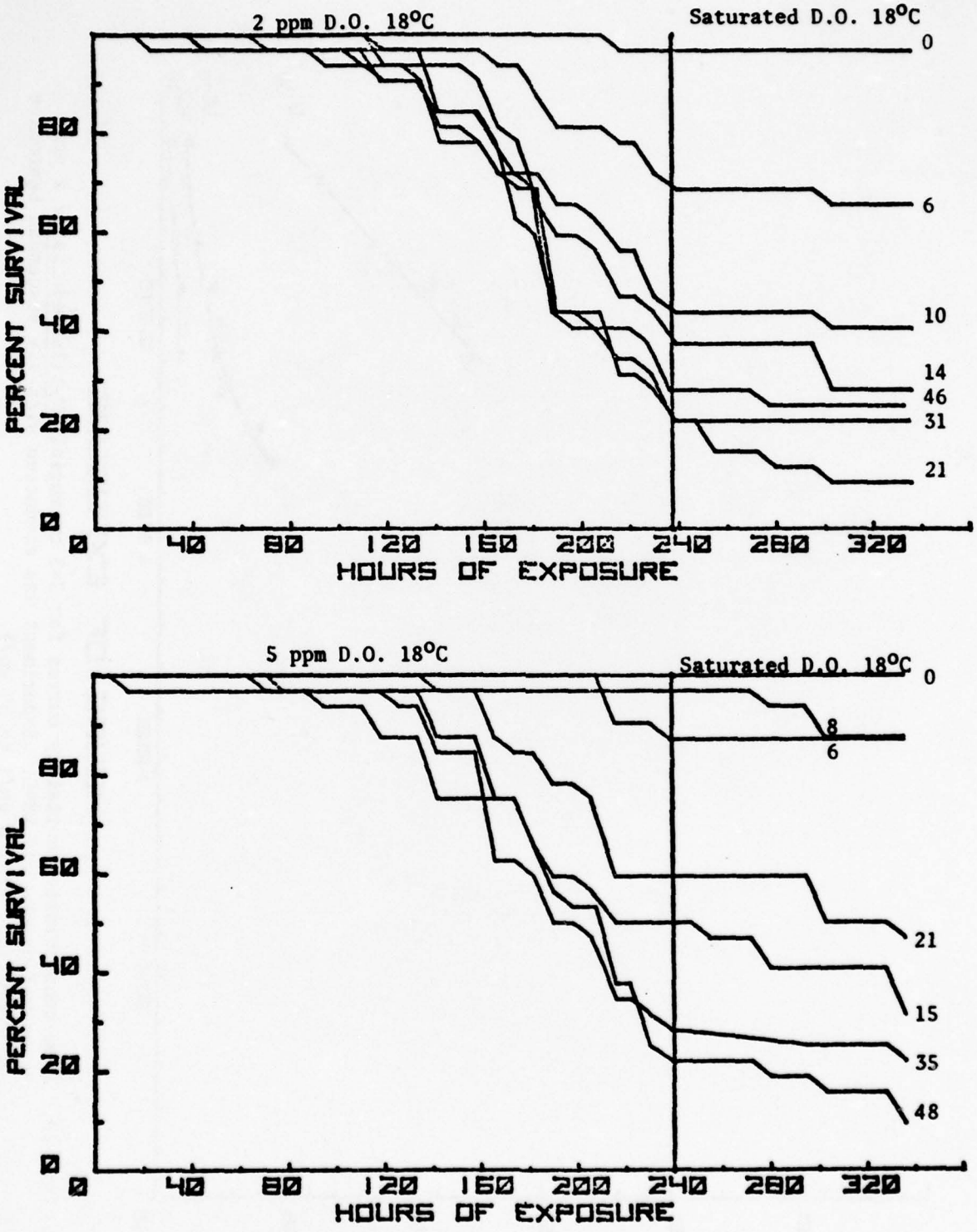


Fig. 20. Percent survival of 2-3 cm Mytilus edulis during 240 hours of exposure to suspended bentonite and an additional 96 hour post-exposure period with the survivors in clear, oxygen-saturated water. Experiments were conducted at 2 ppm and 5 ppm dissolved oxygen and 18°C. Numbers on the lines are suspended bentonite concentrations in gm/l.

was more pronounced for the shrimp C. nigricauda than for the mussels. At 5 ppm dissolved oxygen, the time-concentration mortality curves (Fig. 21) were nearly level by the end of the test, indicating that few new deaths were occurring with increasing exposure time. In contrast, 100% mortality had occurred in three test aquaria and over 50% in the remaining three after 83 hours exposure at 2 ppm dissolved oxygen. This resulted in erratic and unreliable LC_x estimates, and the analysis was halted at that time (Fig. 22). Thus, while the effect of dissolved oxygen on suspended solids tolerance was obviously great, it could not be fully quantified. None of the surviving shrimp died after 96 hours in clear, oxygen saturated water.

Only a few deaths of S. laticauda occurred at 2 ppm dissolved oxygen and these were poorly correlated with suspended solids concentration (Table VI), giving data too erratic for reliable estimation of LC_x values. More deaths occurred at 5 ppm dissolved oxygen (Table VI), although 50% mortality was not reached. The LC_{20} and LC_{10} curves are plotted in Fig. 23. Of 270 surviving isopods, only seven animals scattered among five concentrations died during the post-exposure period.

The experiment with the polychaete N. succinea showed no correlation of increasing suspended solids concentration with mortality. Control mortalities were fairly high, and while more deaths occurred in the test aquaria, they followed no discernible pattern (Table VI).

No M. saxatilis mortalities occurred at 5 ppm dissolved oxygen in suspended bentonite concentrations from 0.2 to 2.0 gm/l (Table VI), and C. aggregata did not suffer 50% mortality under these conditions.

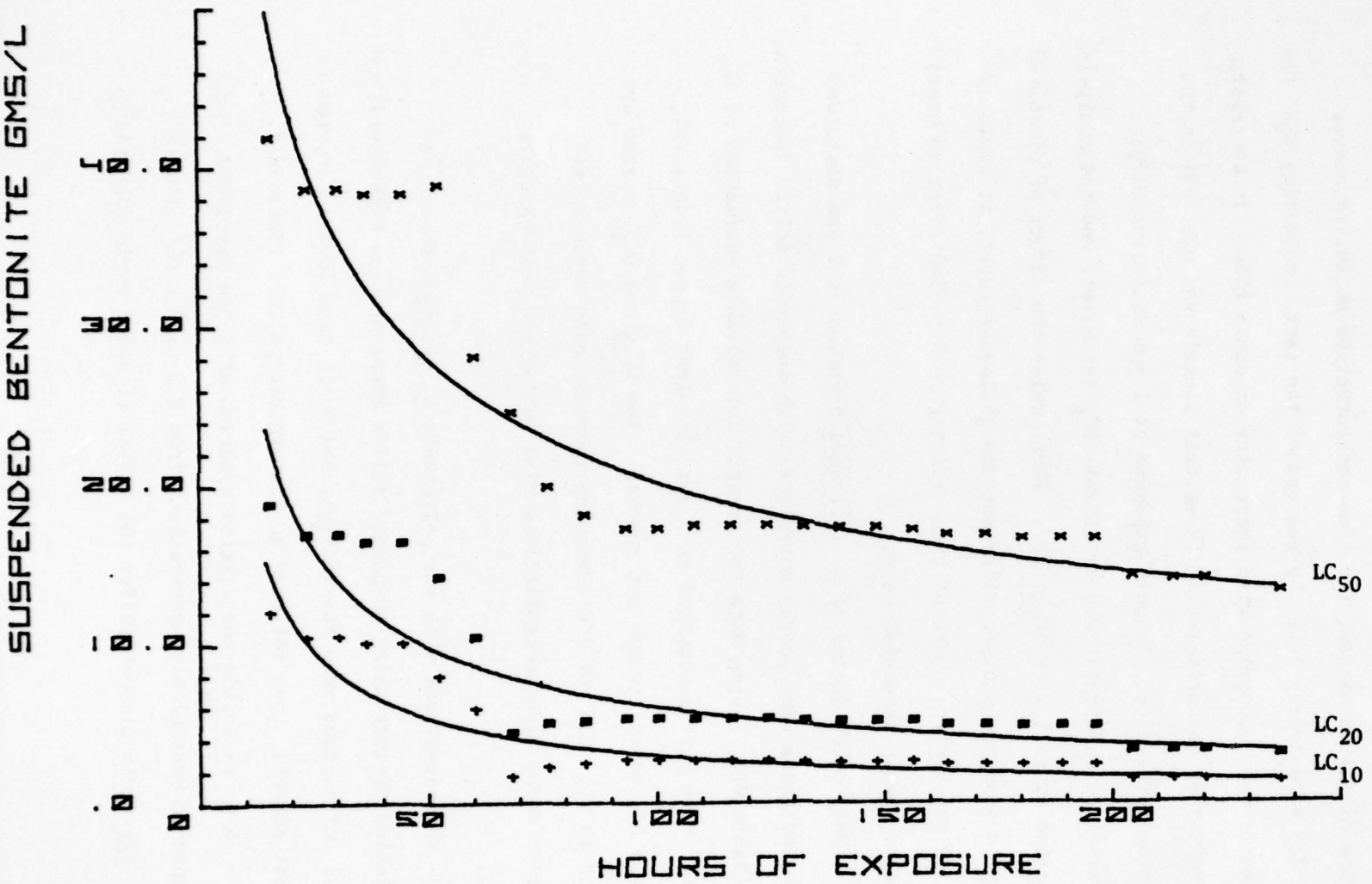


Fig. 21. Time-concentration mortality curves for 3-4 cm sand shrimp Crangon nigricauda at 5 ppm dissolved oxygen and 18°C. Experiment was conducted with six suspended bentonite concentrations from 6 gm/1 to 60 gm/1.

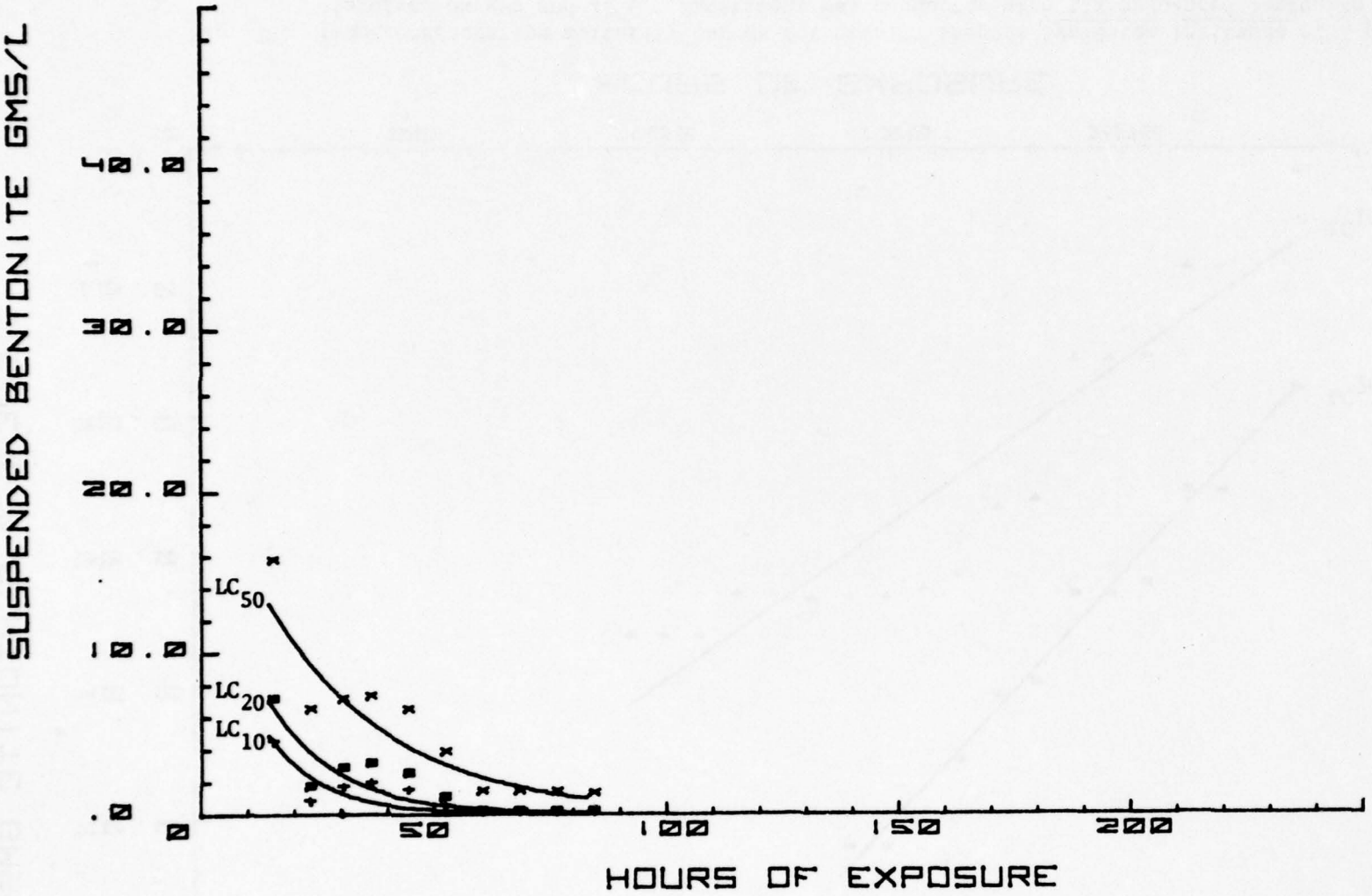


Fig. 22. Time-concentration mortality curves for 3-4 cm sand shrimp Crangon nigricauda at 2 ppm dissolved oxygen and 18°C. Experiment was conducted with six suspended bentonite concentrations from 6 gm/l to 60 gm/l.

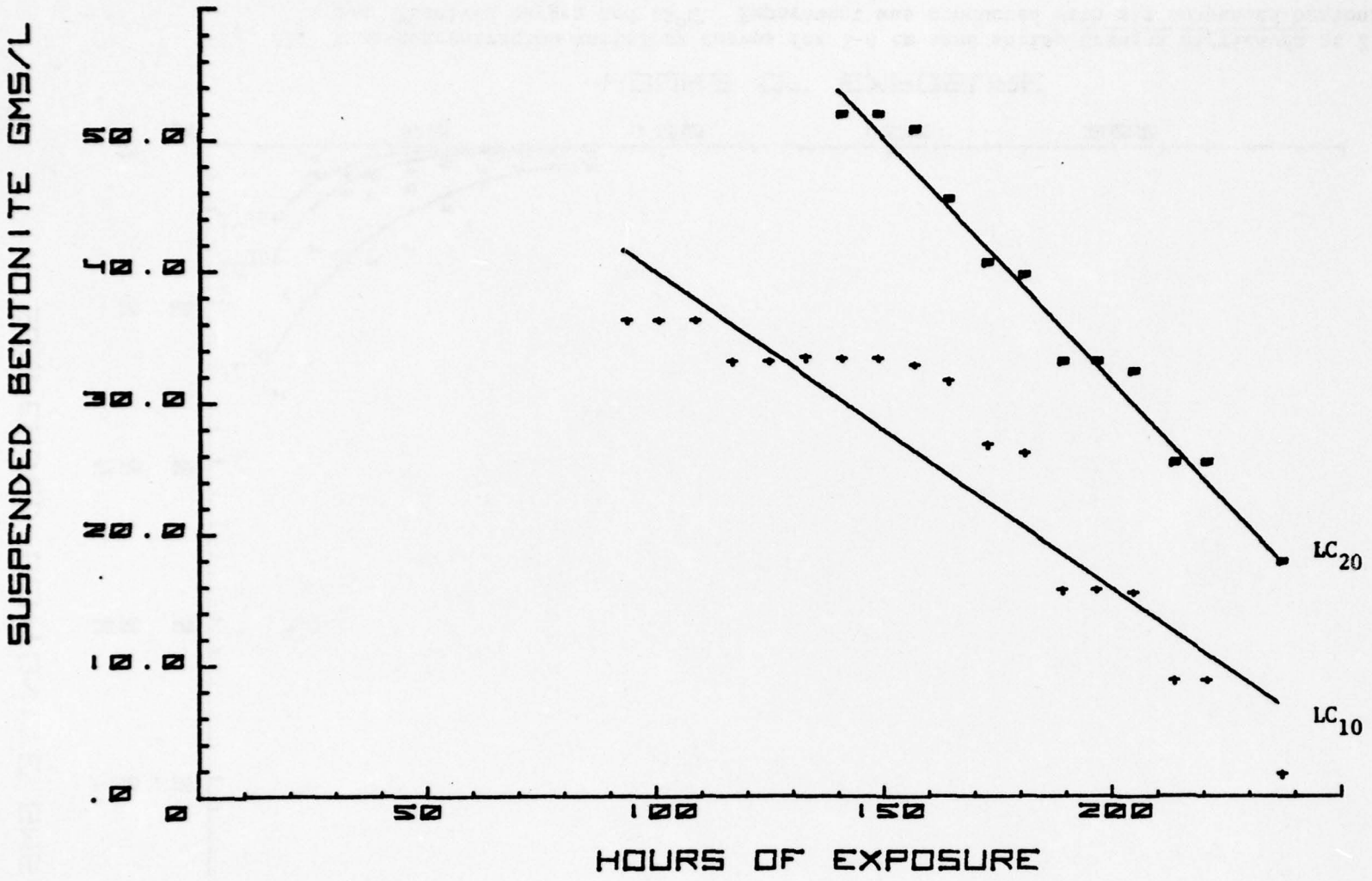


Fig. 23. Time-concentration mortality curves for "adult" isopods Synidotea laticauda at 5 ppm dissolved oxygen and 18°C. Experiment was conducted with six suspended bentonite concentrations from 6 gm/1 to 60 gm/1.

The LC_{20} and LC_{10} curves for this species are plotted in Fig. 24, showing 10% mortality at less than 1 gm/l after 10 days. A suspended solids concentration range of 0.6 gm/l to 6.0 gm/l was used in the 2 ppm dissolved oxygen experiment with both species, producing higher mortalities (Table VI). Deaths occurred rapidly among the M. saxatilis and had stabilized by 100 hours of exposure (Fig. 25). After 70 hours of exposure C. aggregata had greater than 20% mortality in all but one test condition, and the LC_{20} and LC_{10} calculations were halted at that time. The LC_{50} curve is plotted to the end of the experiment in Fig. 26. None of the surviving M. saxatilis died during the post-exposure period, nor did any C. aggregata after testing at 5 ppm dissolved oxygen. However, after testing at 2 ppm dissolved oxygen, the only survivor which had been exposed to 1.3 gm/l and all four of the survivors exposed to 0.9 gm/l died in 96 hours. All the survivors from the controls and 0.6 gm/l lived through the post-exposure period.

Influence of Simultaneous Variations in Temperature and Dissolved Oxygen on the Lethal Effects of Suspended Bentonite

These experiments, conducted at 10°C and 18°C and 2 ppm and 5 ppm dissolved oxygen, utilized a control and three suspended solids concentrations nominally spanning one order of magnitude. The clear water control aquaria were maintained at reduced oxygen levels corresponding to the test aquaria. The conditions under which the experimental organisms were collected, held in the laboratory and tested are presented in Appendix D. Table DII identifies those species tested simultaneously in the same aquaria. The raw data from the

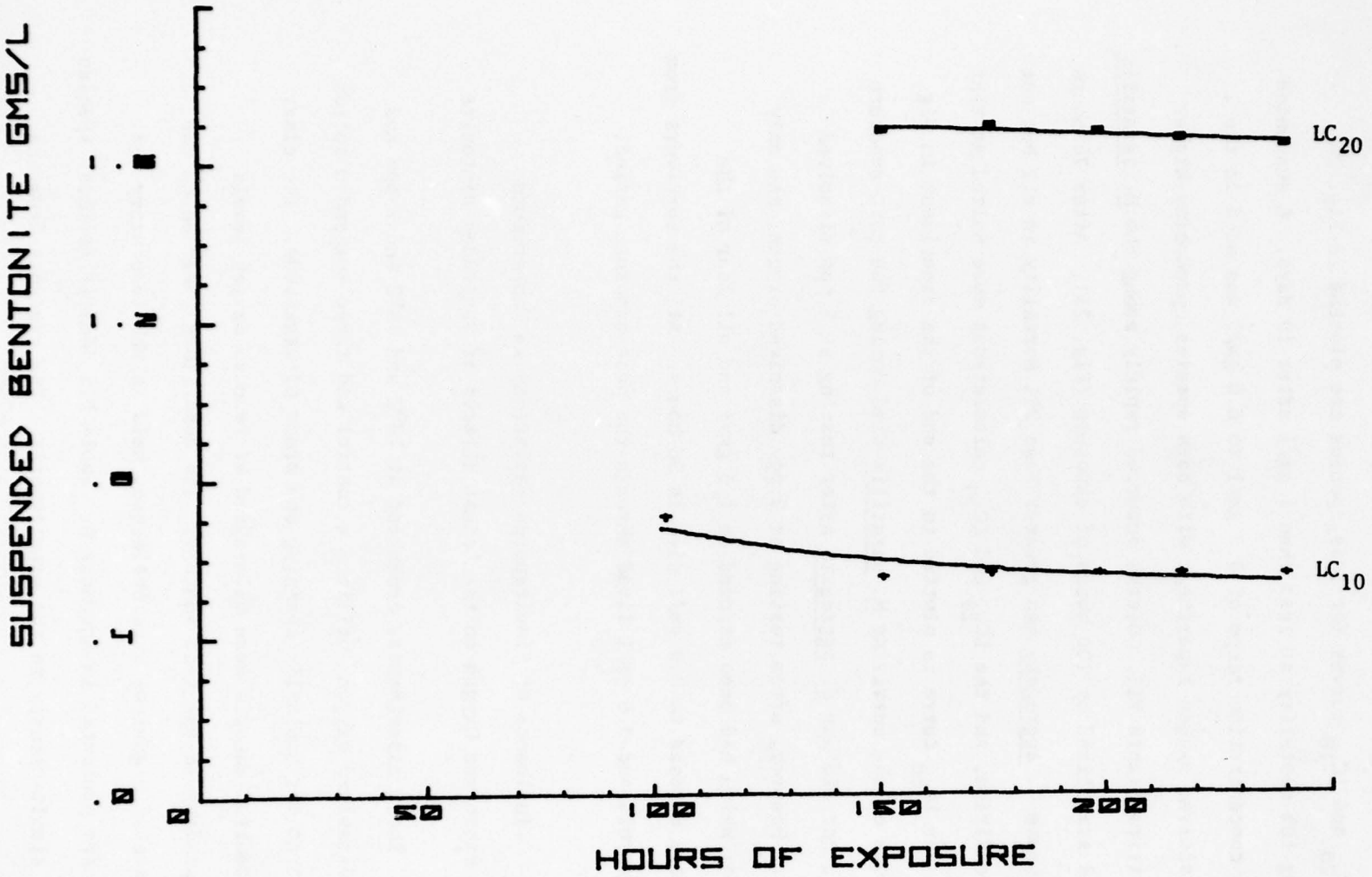


Fig. 24. Time-concentration mortality curves for 5.5-7 cm shiner perch Cymatogaster aggregata at 5 ppm dissolved oxygen and 18°C. Experiment was conducted with six suspended bentonite concentrations from 0.2 gm/1 to 2.0 gm/1.

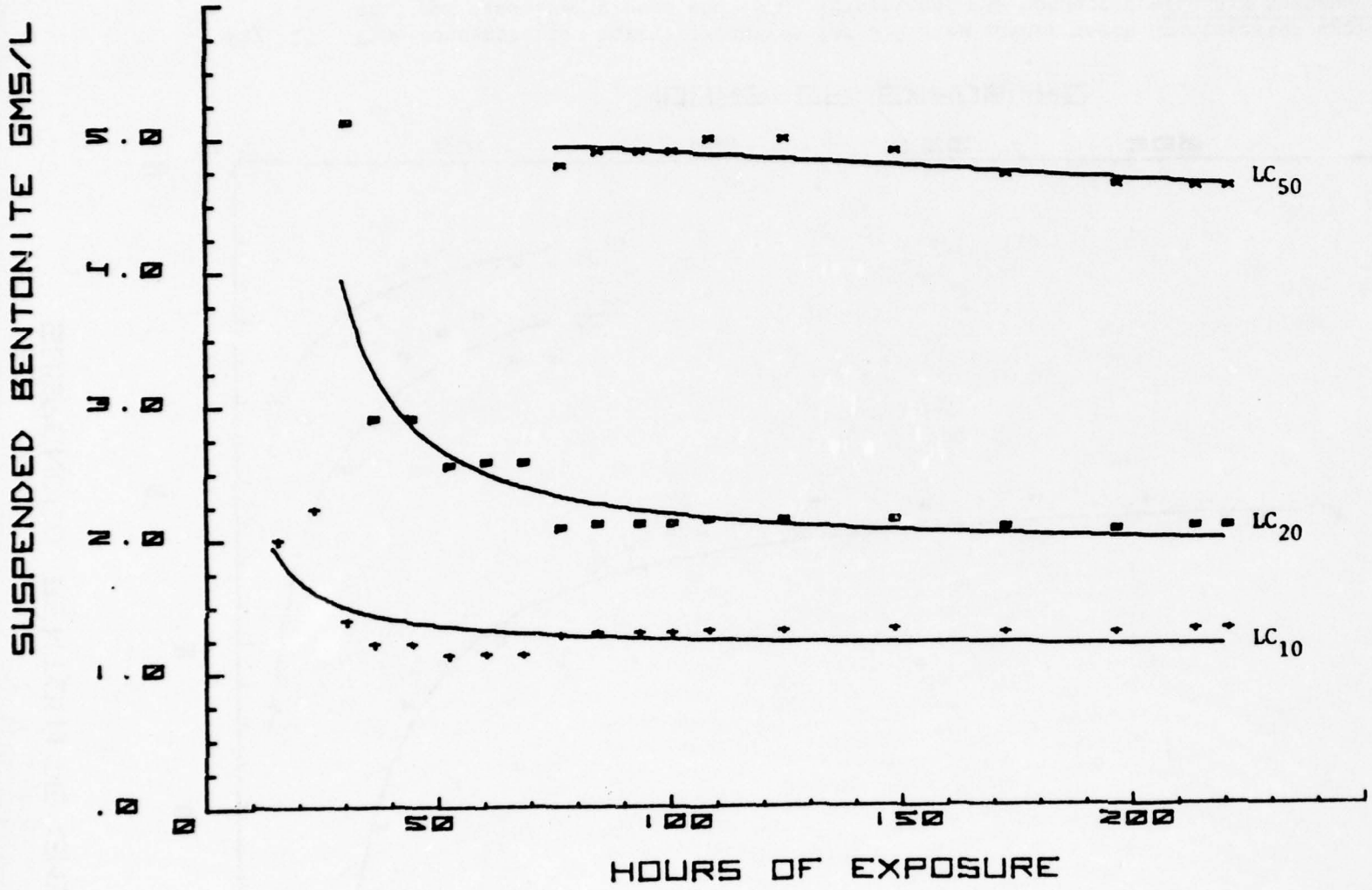


Fig. 25. Time-concentration mortality curves for 5-8 cm striped bass Morone saxatilis at 2 ppm dissolved oxygen and 18°C. Experiment was conducted with six suspended bentonite concentrations from 0.6 gm/1 to 6.0 gm/1.

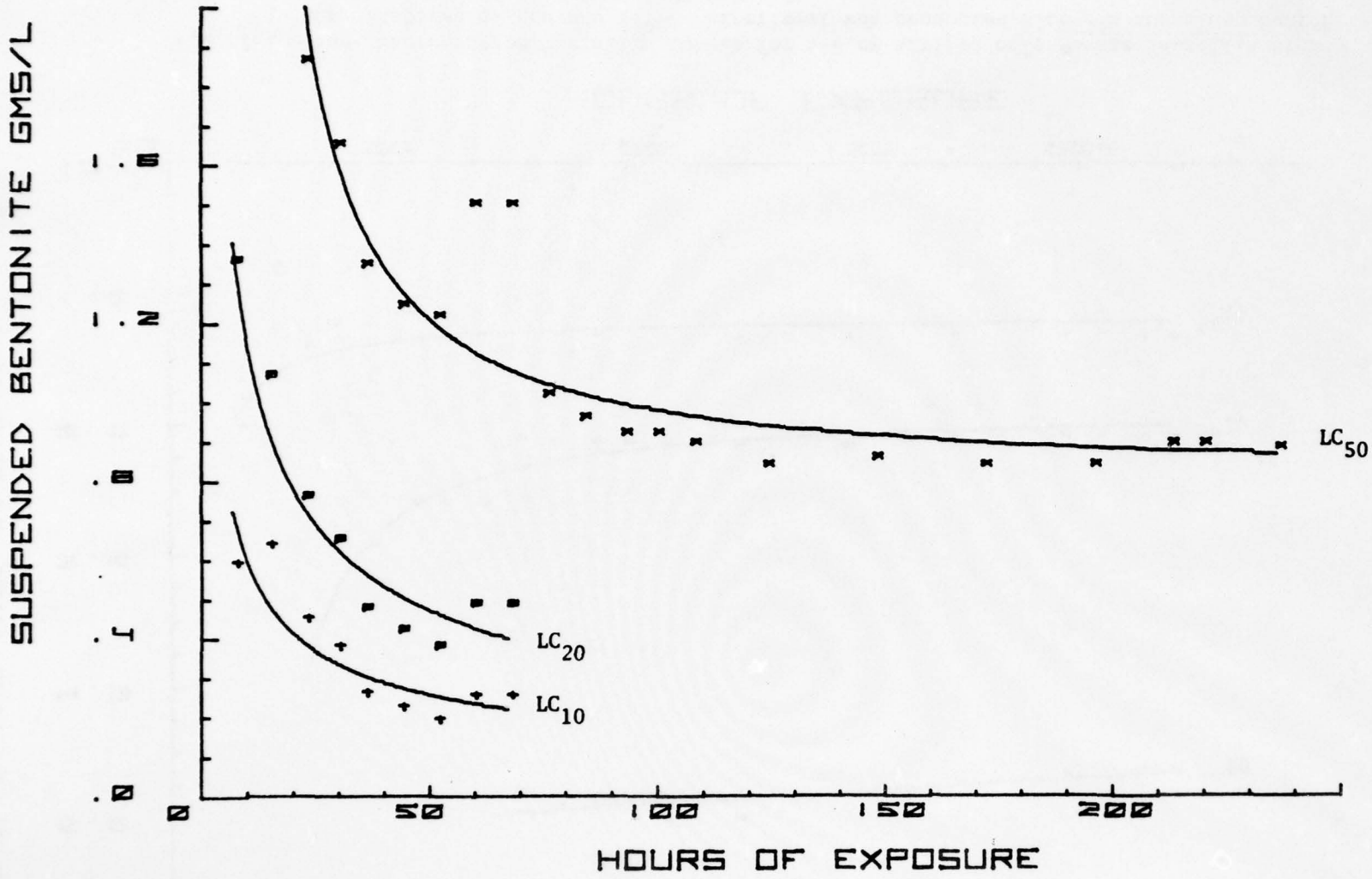


Fig. 26. Time-concentration mortality curves for 5.5-7 cm shiner perch Cymatogaster aggregata at 2 ppm dissolved oxygen and 18°C. Experiment was conducted with six suspended bentonite concentrations from 0.6 gm/l to 6.0 gm/l.

multifactor experiments are summarized in Table VIII.

The survival of M. edulis over time is illustrated in Fig. 27 for two dissolved oxygen levels and four suspended bentonite concentrations at 18°C, and in Fig. 28 for the same conditions at 10°C. Figures 27 and 28 indicate that survival was greater at 10°C than at 18°C, but reveal little difference related to dissolved oxygen. The results of the factorial analysis of variance comparing individual times of survival under each test condition are presented in Table IX. This confirmed that the effects of dissolved oxygen levels on length of survival were not significant, nor were any of the interaction terms involving dissolved oxygen. The effects due to both suspended bentonite concentration and temperature were highly significant, as was the bentonite-temperature interaction. This indicated that these factors interacted in a non-additive manner to influence survival time, and that the effects of changes in suspended bentonite concentration depended on temperature. For example, it was not possible to specify the mean survival time at 8 gm/l unless the statement was limited to a particular temperature.

Because of this interdependence of factors the data were re-analyzed using a one-way analysis of variance with the eight combinations of suspended bentonite concentration and temperature regarded as separate treatments. This analysis, and the mean contrast based upon it, are presented in Table X. The mean contrast revealed no significant differences among the responses to any suspended bentonite concentration at 10°C, although the length of survival tended to vary inversely with concentration. The response was different at 18°C, with significant

Table VIII

Summary of raw data from mortality tests with suspended bentonite at 2 ppm and 5 ppm dissolved oxygen and 10°C and 18°C, showing species, approximate size of the animals, experimental temperature and dissolved oxygen levels, length of exposure to test conditions, and the suspended solids concentration, total number of deaths and original number of animals in each experimental condition.

				Suspended Bentonite Concentrations											
				1			2			3			4		
Species	Temp °C	D.O. ppm	Hours of Exp.	gm/l	No. Dead	Orig. No.	gm/l	No. Dead	Orig. No.	gm/l	No. Dead	Orig. No.	gm/l	No. Dead	Orig. No.
Mytilus edulis 2-3 cm	18	5	237	0	1	14	11	5	15	23	12	16	51	15	16
	18	2	237	0	1	16	7	11	16	20	12	16	54	15	15
	10	5	237	0	0	16	7	2	16	15	2	16	52	3	16
	10	2	237	0	1	16	8	5	16	19	4	16	48	3	15
Crangon nigricauda 3-5 cm	18	5	244	0	4	15	9	0	16	17	5	16	44	12	16
	18	2	244	0	6	16	9	16	16	22	15	15	47	16	16
	10	5	244	0	3	16	9	3	14	18	4	16	45	5	16
	10	2	244	0	5	16	7	15	15	23	16	16	46	16	16
Synidotea laticauda "adult"	18	5	244	0	1	15	9	3	15	17	0	15	44	8	13
	18	2	244	0	4	15	9	1	13	22	3	15	47	5	14
	10	5	244	0	2	15	9	2	14	18	2	13	45	2	13
	10	2	244	0	2	14	7	1	14	23	2	15	46	4	15

Table VIII, Continued

Species	Temp °C	D.O. ppm	Hours of Exp.	Suspended Bentonite Concentrations 1 gm/l	1 No. Dead	1 Orig. No.	2 gm/l	2 No. Dead	2 Orig. No.	3 gm/l	3 No. Dead	3 Orig. No.	4 gm/l	4 No. Dead	4 Orig. No.
Morone saxatilis 5.5 - 8 cm	18	5	238	0	0	15	0.6	0	15	1.9	0	15	6.0	2	15
	18	2	238	0	0	15	0.7	2	15	1.9	12	15	7.1	15	15
	10	5	238	0	0	15	0.9	0	15	2.9	8	15	5.4	15	15
	10	2	238	0	0	15	0.6	5	15	1.6	15	15	6.6	15	15
Cymatogaster aggregata 5.5-7.5 cm	18	5	238	0	0	15	0.6	1	15	1.9	5	15	6.0	15	15
	18	2	238	0	1	15	0.7	11	15	1.9	15	15	7.1	15	15
	10	5	238	0	1	15	0.9	0	15	2.9	15	15	5.4	15	15
	10	2	238	0	1	15	0.6	9	15	1.6	15	15	6.6	15	15

Table IX

Factorial analysis of variance table for the multifactor mortality experiment with Mytilus edulis. Each treatment cell contained 14 observations of hours of survival in a 240-hour experiment.

Source	df	SS	MS	F
Suspended bentonite concentration	3	101188	33729	17.7**
Temperature	1	87729	87729	46.0**
Dissolved oxygen	1	299	299	0.2 ns
Suspended bentonite-temperature interaction	3	34691	11564	6.1**
Suspended bentonite-dissolved oxygen interaction	3	11165	3722	1.9 ns
Temperature-dissolved oxygen interaction	1	446	446	0.2 ns
Suspended bentonite-temperature-dissolved oxygen interaction	3	6279	2093	1.1 ns
Error	208	396614	1907	

ns: not significant p .05

**: p<.01

Table X

One-way analysis of variance table and mean contrast for the comparison of the hours of survival in a 240-hour test of Mytilus edulis under the 8 interdependent combinations of suspended bentonite concentration and temperature. In the mean contrast those means underlined by the same line are not different at the .05 probability level, while those not joined by the same line are significantly different.

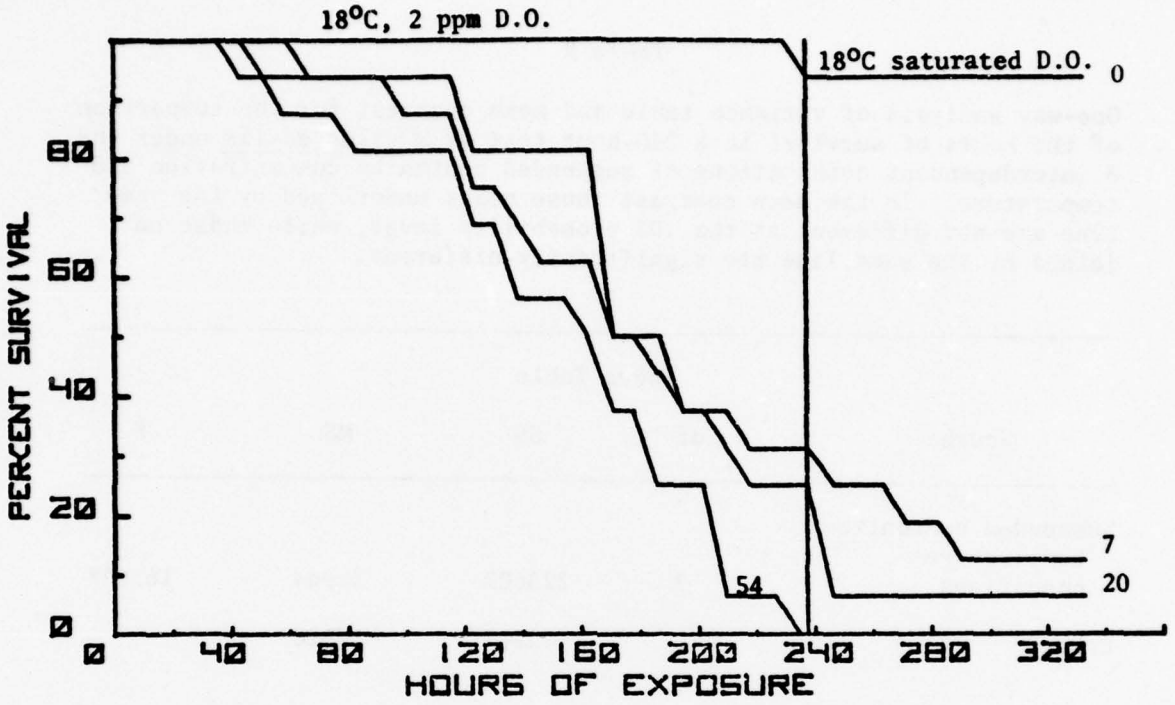
Anova Table

Source	df	SS	MS	F
Suspended bentonite-temperature conditions	7	223609	31944	16.6**
Error	216	414803	1920	

**p<.01

Mean Contrast

Suspended bentonite-gm/l	51	19	8	51	19	8	0	0
Temperature - C^{o}	18	18	18	10	10	10	18	10
Mean Survival time - hours	151	167	193	216	223	230	240	240



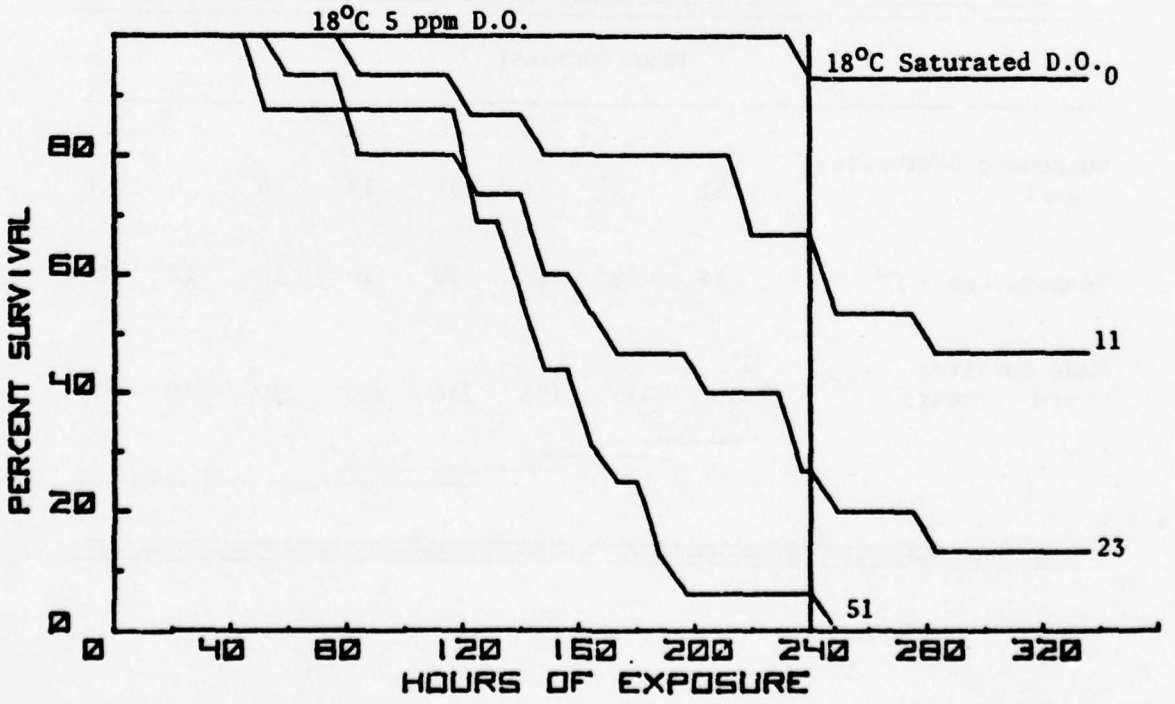


Fig. 27. Percent survival of 2-3 cm Mytilus edulis during 240 hours of exposure to suspended bentonite and an additional 96-hour post-exposure period with the survivors in clear, oxygen-saturated water. This phase of the experiment was conducted at 2 ppm and 5 ppm dissolved oxygen and 18°C. Numbers on the lines are suspended bentonite concentrations in gm/l.

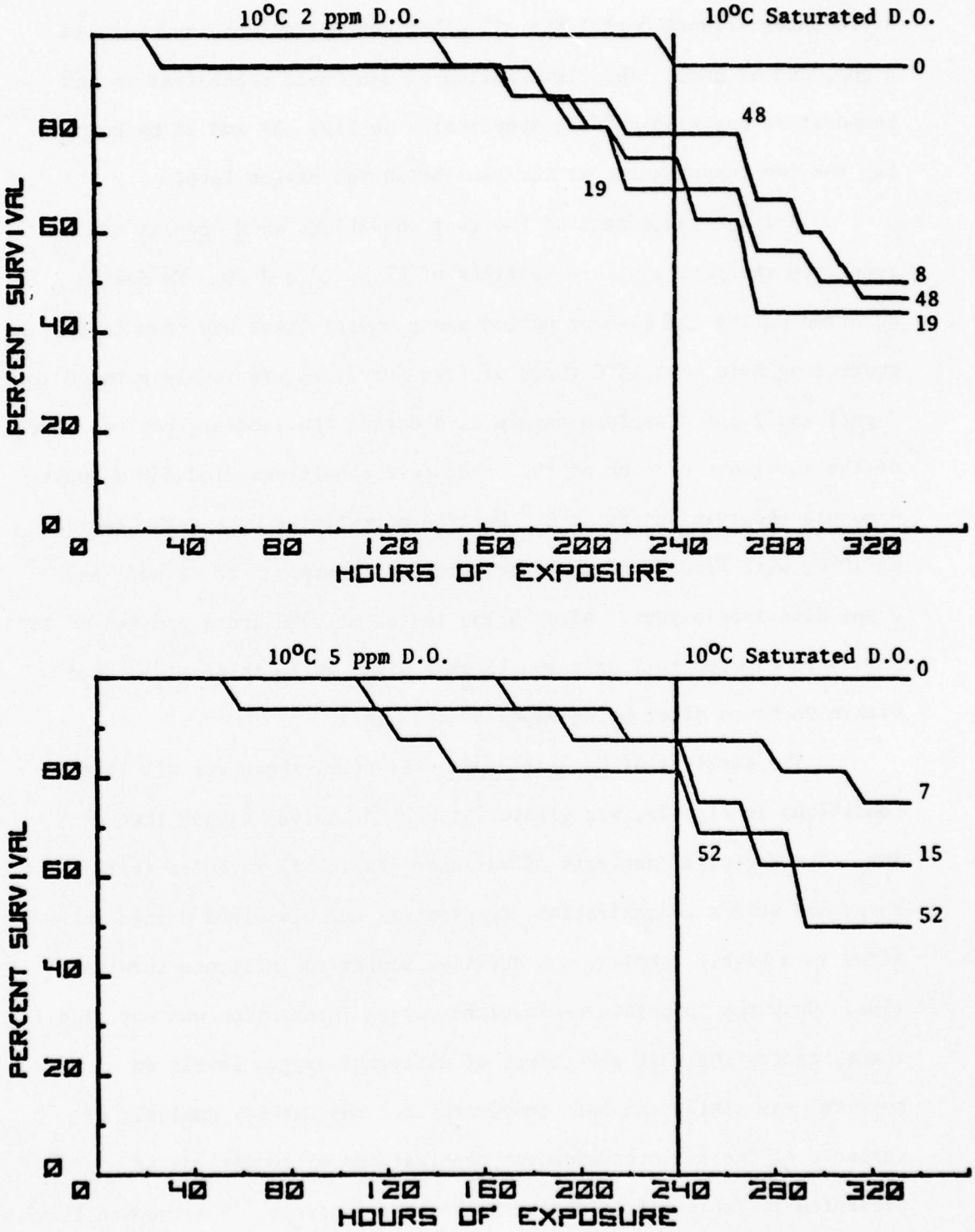


Fig. 28. Percent survival of 2-3 cm Mytilus edulis during 240 hours of exposure to suspended bentonite and an additional 96-hour post-exposure period with the survivors in clear, oxygen-saturated water. This phase of the experiment was conducted at 2 ppm and 5 ppm dissolved oxygen at 10°C. Numbers on the lines are suspended bentonite concentrations in gm/l.

differences between 0 gm/l and all other concentrations, and between 8 gm/l and 51 gm/l. This interaction of bentonite concentration and temperature can also be seen graphically in Figs. 27 and 28 by comparing the two temperatures at the same dissolved oxygen level.

A lingering effect of the test conditions on M. edulis can be seen from the post-exposure sections of Figs. 27 and 28. No deaths occurred during the 96-hour period among mussels that had been in the control aquaria. At 18°C three of five survivors previously exposed to 7 gm/l and 2 ppm dissolved oxygen died during the recovery period. One of the survivors of each of the other test conditions died during post-exposure observations at 18°C. Delayed mortalities were more numerous at 10°C, with five of 12 survivors previously exposed to 48 gm/l and 2 ppm dissolved oxygen. After being tested at 10°C and 5 ppm two of 14 survivors exposed to 7 gm/l and 15 gm/l died, while three of 13 died within 96 hours after being exposed to 52 gm/l.

The survival of C. nigricauda over time, shown for all test conditions in Fig. 29, was greater at high dissolved oxygen than at low. The factorial analysis of variance (Table XI) revealed that suspended solids concentration, temperature and dissolved oxygen interacted in a highly complex, non-additive manner to influence survival time. Only the temperature-dissolved oxygen interaction was not significant, indicating that the effect of different oxygen levels on survival was similar at both temperatures. The one-way analysis of variance of the 16 interdependent combinations of conditions is presented in Table XII, along with the mean contrast. This showed that survival was lowest at the higher suspended bentonite and low dissolved

Table XI

Factorial analysis of variance table for the multifactor experiment with Crangon nigricauda. Each treatment cell contained 14 observations of hours of survival in a 240-hour experiment.

Source	df	SS	MS	F
Suspended bentonite concentration	3	425854	141951	35.3**
Temperature	1	16201	16201	4.0*
Dissolved oxygen	1	658161	658161	163.8**
Suspended bentonite-temperature interaction	3	82937	27646	6.9**
Suspended bentonite-dissolved oxygen interaction	3	122412	40804	10.2**
Temperature-dissolved oxygen interaction	1	1507	1507	0.4 ns
Suspended bentonite-temperature-dissolved oxygen interaction	3	39422	13140	3.3*
Error	208	835705	4018	

ns: not significant p .05

*: $p<.05$

**: $p<.01$

Table XII

One-way analysis of variance table and mean contrast for the comparison of the hours of survival in a 240-hour test of Crangon nigricauda under the 16 interdependent combinations of suspended bentonite concentration, temperature and dissolved oxygen. In the mean contrast those means underlined by the same line are not different at the .05 probability level, while those not joined by the same line are significantly different.

Anova Table

Source	df	SS	MS	F
Suspended bentonite-temperature-dissolved oxygen conditions	15	1346496	89766	22.3**
Error	208	835705	4018	

** p<.01

Mean Contrast

Suspended bentonite-gm/l	45	45	20	9	20	45	9	0	9	20	45	0	20	0	0	9
Temperature-^{o}C	18	10	18	10	10	18	18	18	10	18	10	18	10	10	10	18
Dissolved oxygen-ppm	2	2	2	2	2	5	2	2	5	5	5	5	5	2	5	5
Mean survival time-hours	13	14	44	65	99	108	119	170	196	203	205	209	214	216	234	240

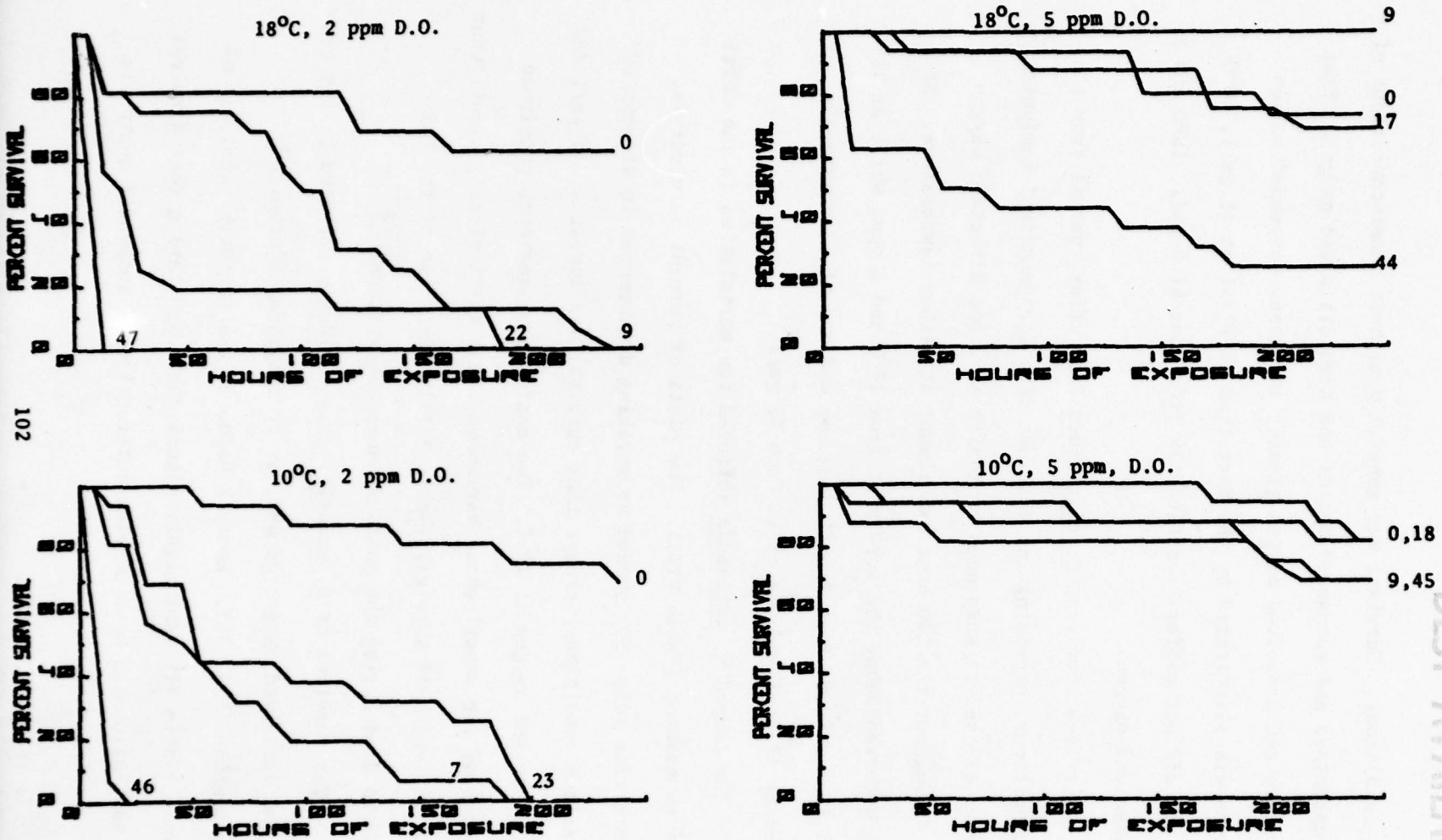


Fig. 29. Percent survival of 3-5 cm Crangon nigricauda during 240 hours of exposure to suspended bentonite. The experiment was conducted at 10°C and 18°C and 2 ppm and 5 ppm dissolved oxygen. Numbers on the lines are suspended bentonite concentrations in gm/l.

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oxygen conditions. Survival was generally highest under conditions of low temperature and suspended solids and high dissolved oxygen. That temperature and dissolved oxygen greatly influence suspended solids tolerance was illustrated by the fact that survival at 45 gm/l, 10°C and 5 ppm did not differ significantly from that at 0 gm/l, 18°C and 2 ppm dissolved oxygen.

Very few surviving C. nigricauda died after removal from the test conditions, providing insufficient data for graphical analysis. No shrimp exposed to suspended bentonite at 2 ppm dissolved oxygen lived throughout the 240-hour experiment at either temperature. No deaths occurred among the survivors from 18°C and 5 ppm, while at 10°C two of 13 survivors from 9 gm/l died, as did one of 13 previously exposed to 18 gm/l and one of 11 from 45 gm/l.

The isopod S. laticauda suffered few mortalities in the multi-factor experiment (Table VIII). The plots of percent survival vs. exposure time (Fig. 30) showed no striking differences in effects of the various conditions, except that survival was lowest at 44 gm/l and 5 ppm dissolved oxygen at 18°C. The analysis of variance confirmed that none of the experimental variables had a statistically significant effect on length of survival (Table XIII). Only two of the 187 survivors died during the post-exposure observations.

The survival of M. saxatilis was obviously affected by temperature and dissolved oxygen as well as by suspended bentonite concentration (Fig. 31). Every F value in the factorial analysis of variance (Table XIV) was highly significant, indicating that survival time was influenced by a complex interaction of suspended bentonite

Table XIII

Factorial analysis of variance table for the multifactor experiment with Synidotea laticauda. Each treatment cell contained 13 observations of hours of survival in a 240-hour experiment.

Source	df	SS	MS	F
Suspended bentonite concentration	1	13160	4387	1.1 ns
Temperature	1	3360	3360	0.9 ns
Dissolved oxygen	1	208	208	0.1 ns
Suspended bentonite-temperature interaction	3	23261	7754	2.0 ns
Suspended bentonite-dissolved oxygen interaction	3	6969	2324	0.6 ns
Temperature-dissolved oxygen interaction	1	29	29	0.1 ns
Suspended bentonite-temperature-dissolved oxygen interaction	3	9154	3053	0.8 ns
Error	192	737256	3840	

ns: not significant p .05

Table XIV

Factorial analysis of variance table for the multifactor experiment with Morone saxatilis. Each treatment cell contained 15 observations of hours of survival in a 240-hour experiment.

Source	df	SS	MS	F
Suspended bentonite concentration	3	1162049	387350	176.7**
Temperature	1	113666	113666	51.9**
Dissolved oxygen	1	353741	353741	161.4**
Suspended bentonite-temperature interaction	3	65951	21984	10.0**
Suspended bentonite-dissolved oxygen interaction	3	210419	70140	32.0**
Temperature-dissolved oxygen interaction	1	32713	32713	14.9**
Suspended bentonite-temperature-dissolved oxygen interaction	3	107513	35838	16.3**
Error	224	490991	2192	

** $p<.01$

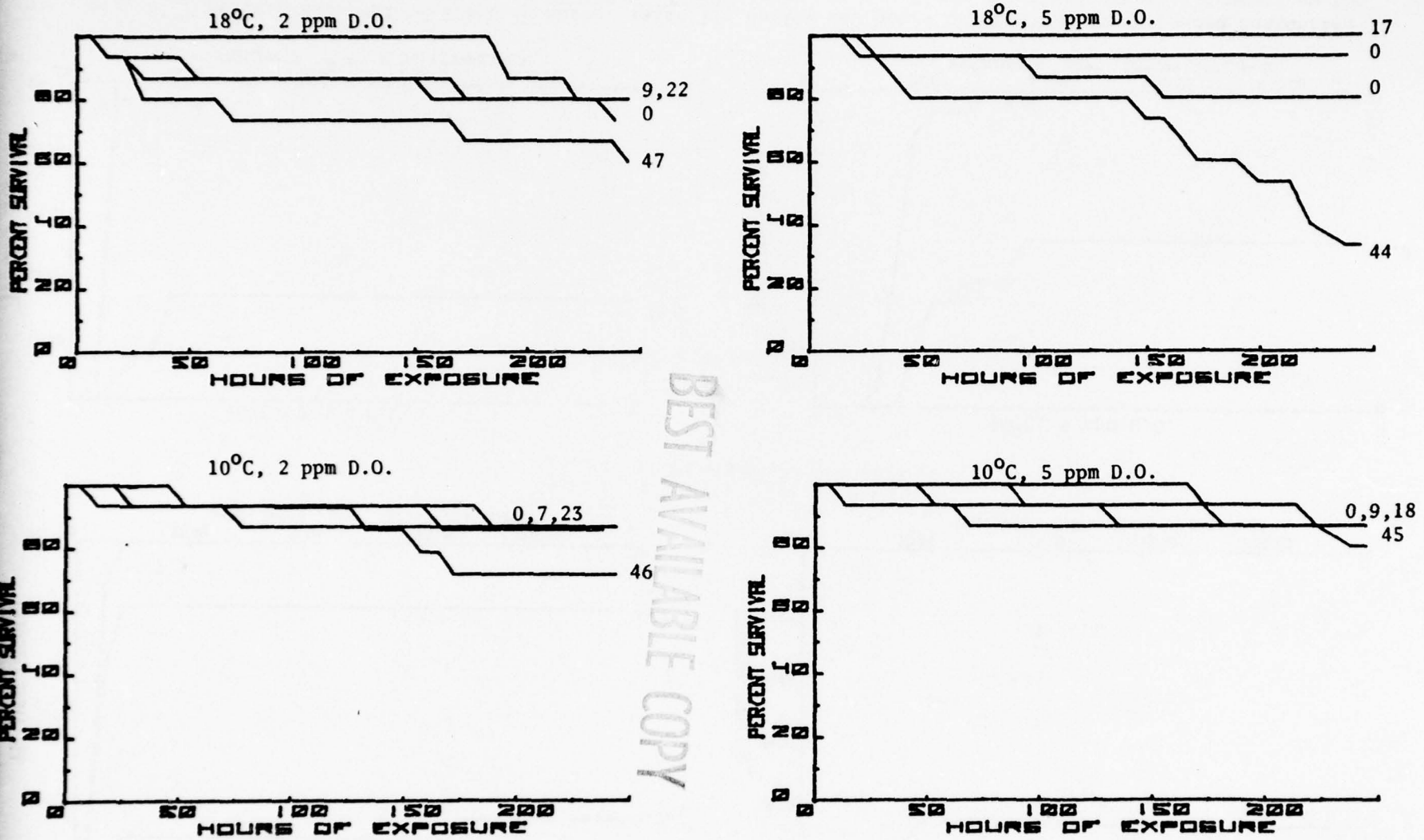

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Fig. 30. Percent survival of "adult" Synidotea laticauda during 240 hours of exposure to suspended bentonite. The experiment was conducted at 10°C and 18°C and 2 ppm and 5 ppm dissolved oxygen. Numbers on the lines are suspended bentonite concentrations in gm/l.

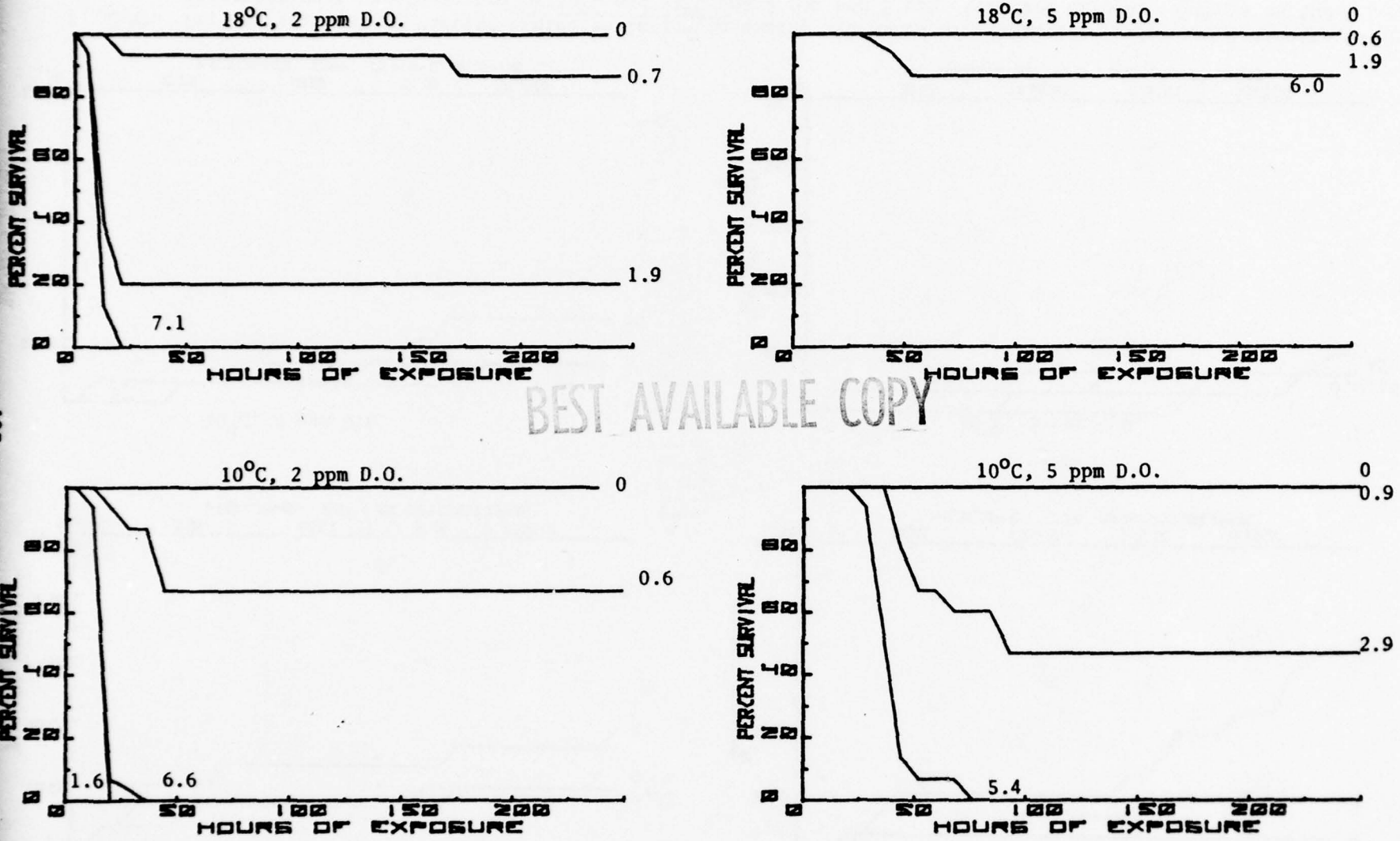

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Fig. 31. Percent survival of 5.5-8 cm Morone saxatilis during 240 hours of exposure to suspended bentonite. The experiment was conducted at 10°C and 18°C and 2 ppm and 5 ppm dissolved oxygen. Numbers on the lines are suspended bentonite concentrations in gm/l.

concentration, temperature and dissolved oxygen. The one-way analysis of variance of the 16 combinations of these factors is presented in Table XV. The mean contrast showed no deaths in the control aquaria under any combination of temperature and dissolved oxygen conditions. There were no statistically significant differences among the responses to all suspended bentonite concentrations at 18°C and 5 ppm dissolved oxygen. For every combination of suspended bentonite and dissolved oxygen, survival was longer at 18°C than at 10°C. The lowest survival was generally at conditions of higher concentrations, low oxygen and low temperature. No survivors died during the post-exposure period.

The multifactor experiment confirmed C. aggregata as the most sensitive species tested. The plots of percent survival vs. time (Fig. 32) indicated shorter survival at low temperature and low dissolved oxygen conditions. All interactions of the experimental variables were shown by the factorial analysis of variance to have a highly significant effect on the time of survival (Table XVI). The one-way analysis of variance considering each test condition as a separate treatment is presented in Table XVII, together with the mean contrast. This revealed four significantly different groups of survival times. The lowest group contained the higher concentration and low temperature and dissolved oxygen conditions. The only 18°C, 5 ppm condition included was at 6.4 gm/l. Survival was significantly greater at 0.8 gm/l and 18°C, and greater again at 0.8 gm/l, 10°C, despite only 2 ppm dissolved oxygen at both conditions. The longest survival was in the controls under all conditions. Also included in the group not different from the controls was the survival at 0.8 gm/l and 5 ppm at both temperatures,

Table XV

One-way analysis of variance table and mean contrast for the comparison of the hours of survival in a 240-hour test of Morone saxatilis under the 16 interdependent combinations of suspended bentonite concentration, temperature and dissolved oxygen. In the mean contrast those means underlined by the same line are not different at the .05 probability level, while those not joined by the same line are significantly different.

Anova Table

Source	df	SS	MS	F
Suspended bentonite-temperature-dissolved oxygen conditions	15	2046052	136403	62.2**
Error	224	490991	2192	

** p<.01

Mean contrast

Suspended bentonite-gm/l	6.4	2.2	6.4	6.4	2.2	2.2	0.8	6.4	0.8	2.2	0.8	0.8	0.0	0.0	0.0	0.0
Temperature-°C	18	10	10	10	18	10	10	18	18	18	10	18	10	18	10	18
Dissolved oxygen-ppm	2	2	2	5	2	5	2	5	2	5	5	5	2	2	5	5
Mean survival time-hours	14	20	20	42	60	145	173	215	221	240	240	240	240	240	240	240

Table XVI

Factorial analysis of variance table for the multifactor experiment with Cymatogaster aggregata. Each treatment cell contained 15 observations of hours of survival in a 240-hour experiment.

Source	df	SS	MS	F
Suspended bentonite concentration	3	1802763	600921	442.7**
Temperature	1	8178	8178	6.0**
Dissolved oxygen	1	209214	209214	154.1**
Suspended bentonite-temperature interaction	3	111739	37246	27.4**
Suspended bentonite-dissolved oxygen interaction	3	139594	46531	34.3**
Temperature-dissolved oxygen interaction	1	50297	50297	38.4**
Suspended bentonite-temperature-dissolved oxygen interaction	3	70235	23412	17.2**
Error	224	304037	1357	

** $p<.01$

Table XVII

One-way analysis of variance table and mean contrast for the comparison of the hours of survival in a 240-hour test of Cymatogaster aggregata under the 16 interdependent combinations of suspended bentonite concentration, temperature and dissolved oxygen. In the mean contrast those means underlined by the same line are not different at the .05 probability level, while those not joined by the same line are significantly different.

Anova Table

Source	df	SS	MS	F
Suspended bentonite-temperature-dissolved oxygen conditions	15	2393821	159588	117.6**
Error	224	304037	1357	

**p<.01

Mean Contrast

Suspended bentonite-gm/l	6.4	6.4	2.2	2.2	6.4	6.4	2.2	0.8	0.8	2.2	0.0	0.0	0.0	0.8	0.8	0.0
Temperature-^{o}C	18	10	18	10	18	10	10	18	10	18	18	10	10	18	10	18
Dissolved oxygen-ppm	2	2	2	2	5	5	5	2	2	5	2	2	5	5	5	5
Mean Survival time-hours	7	15	15	17	21	28	41	105	162	211	228	231	236	237	240	240
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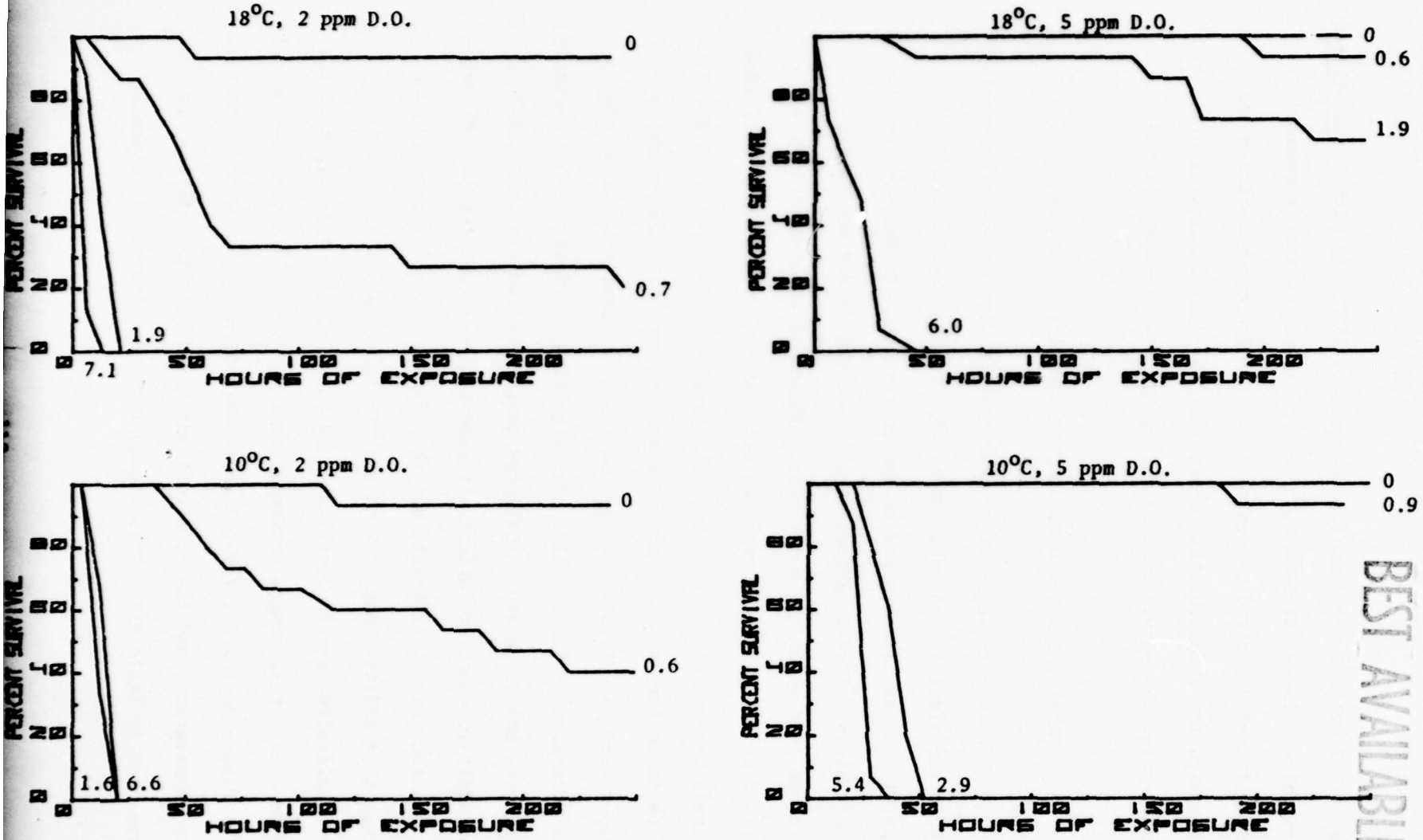

Fig. 32. Percent survival of 5.5-7.5 cm Cymatogaster aggregata during 240 hours of exposure to suspended bentonite. The experiment was conducted at 10°C and 18°C and 2 ppm and 5 ppm dissolved oxygen. Numbers on the lines are suspended bentonite concentrations in gm/l.

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and at 2.2 gm/l, 5 ppm dissolved oxygen and 18°C. As with M. saxatilis, survival was longer at 18°C than at 10°C under the same suspended bentonite and dissolved oxygen conditions. The opposite trend was found with the invertebrates, whose survival varied directly with temperature. No survivors died during the post-exposure period.

Other Responses Noted During Mortality Experiments

The mussels were not allowed time to form byssal attachments before the start of the experiment. By the first observation all control mussels had attached to the containers. In the test aquaria some mussels failed to attach at all, and others did not remain attached throughout the experiment. Therefore attachment time was defined as the period, to the nearest 8 hours, during which individual mussels had byssal attachments. The results of the factorial analysis of variance for the influence of suspended bentonite concentration, temperature and dissolved oxygen on the attachment time of M. edulis are presented in Table XVIII. The significant interaction of suspended bentonite and temperature indicated that these factors combined in a non-additive manner to affect attachment time. The dissolved oxygen main effect was significant, but this factor did not interact in an other than additive manner with the remaining variables. That is, lower dissolved oxygen levels reduced survival time regardless of the temperature or suspended bentonite concentration. The one-way analysis of variance of the 8 inter-related combinations of suspended bentonite and temperature, and the mean contrast based on this analysis, are presented in Table XIX. This showed that the effects of suspended

Table XVIII

Factorial analysis of variance table for the multifactor byssal attachment experiment with Mytilus edulis. Each treatment cell contained 14 observations of hours of byssal attachment of individual mussels in a 240-hour experiment.

Source	df	SS	MS	F
Suspended bentonite concentration	3	1220000	405470	81.3 **
Temperature	1	179723	179723	36.0 **
Dissolved oxygen	1	20349	20349	4.1 *
Suspended bentonite-temperature interaction	3	128701	42900	8.6 **
Suspended bentonite-dissolved oxygen interaction	3	22839	7613	1.5 ns
Temperature-dissolved oxygen interaction	1	3673	3673	0.7 ns
Suspended bentonite-temperature-dissolved oxygen interaction	3	27098	9033	1.8 ns
Error	208	1040000	4989	

ns: not significant p .05

*: $p<.05$

**: $p<.01$

Table XIX

One-way analysis of variance table and mean contrast for the comparison of hours of byssal attachment in a 240-hour test of Mytilus edulis under the 8 interdependent combinations of suspended bentonite concentration and temperature. In the mean contrast those means underlined by the same line are not different at the .05 probability level, while those not joined by the same line are significantly different.

Source	df	SS	MS	F
Suspended bentonite-temperature conditions	7	1524839	217834	42.3**
Error	216	1111734	5147	

** p<.01

Mean Contrast

Suspended bentonite-gm/l	51	51	19	8	19	8	0	0
Temperature-°C	18	10	18	18	10	10	18	10
Mean survival time-hours	22	43	57	90	174	179	240	240

bentonite were greatest at 51 gm/l regardless of temperature. The effects of 19 gm/l and 8 gm/l at 18°C did not differ statistically from those of 51 gm/l at 10°C. Attachment time in 19 gm/l and 8 gm/l at 10°C was significantly greater than for the same concentrations at 18°C. In the controls all mussels remained attached throughout the experiment regardless of temperature. It was subjectively noted that attachment was maintained by progressively fewer and thinner threads before it was abandoned entirely.

There was an indication, even among the small mussels used, of differential mortalities related to size of the organisms. Table XX presents the results of an analysis of variance comparing the length of survival of four size classes of mussels combined from all experimental conditions of all the mussel tests with bentonite. There was a highly significant difference in the times of survival of the size classes. The mean contrast showed significantly shorter survival of mussels less than 15 mm long than with the other size classes, which did not differ among themselves.

Table XXI presents the results of analysis of the time until death of size-classes of C. nigricauda in the suspended bentonite-temperature-dissolved oxygen experiment. Although the differences in survival times were not large enough to produce a significant F value, there was a trend toward longer survival among the smaller shrimp. The mean survival times of each size class and the number of shrimp in each class are presented in Table XXI for inspection.

There was no definite indication that molting was associated with death. In the multifactor experiment a total of 315 shrimp were

Table XX

Analysis of variance and mean contrast comparing the hours of survival of four length classes of Mytilus edulis. Data were compiled from all experimental conditions of all tests of M. edulis with bentonite. The mean contrast was at the .05 probability level. Means joined by the same line are not significantly different while those not joined by the same line differ significantly.

Anova Table

Source	df	SS	MS	F
Treatment (Size classes)	3	35728	11909	4.8**
Error	346	852307	2463	

**p<.01

Mean Contrast

Size class-mm	<15	16-20	26-30	21-25
Number/class	7	65	77	201
Mean-hours	108	169	171	178

Table XXI

Analysis of variance table comparing the hours of survival of four length classes of Crangon nigricauda. Data were compiled from all experimental conditions in the suspended bentonize-temperature-dissolved oxygen mortality experiment. Although the F value was not significant at the .05 probability level, the means are presented for inspection.

Anova Table

Source	df	SS	MS	F
Treatment (Size classes)	2	17929	8964	1.9 ns
Error	126	578721	4593	

ns: not significant p .05

Mean Comparison

Size class-mm	20-29	30-39	40-49
Number/class	14	63	52
Mean survival-hours	95	71	56

tested. Among these a total of 70 molts occurred, 13 (19%) of which the animals failed to survive. These 13 deaths associated with molting were only 8% of the total of 154 deaths occurring. A few shrimp molted twice during the test period.

No differences in survival could be established between ovigerous and non-ovigerous shrimp. Among those that died in the multi-factor experiment, an analysis of variance did not differentiate between the survival times of the two groups.

Analyses of variance showed no evidence at the .05 level of differential mortality related to size with C. aggregata or M. saxatilis.

Effects of Suspended Bentonite on Oxygen Consumption

The conditions under which the experimental animals were collected and held in the laboratory are presented in Appendix E, Table EI, and the test conditions are described in Table EII. The cumulative oxygen consumption during the experiments, which were described on page 31, for all species under each test condition is detailed in Appendix E, Figs. E1-E16. In these figures the individual data points are indicated by + marks, boxes represent means of the replicate observations at each time, the regression estimate is a solid line and the broken lines illustrate the 95% confidence interval around that estimate.

In the absence of animals some settling of bentonite occurred in the respiration containers. A starting concentration of 30 gm/l fell to 14.4 gm/l within 14 hours after the circulating pumps were started (Fig. 33). An original concentration of 13 gm/l decreased to

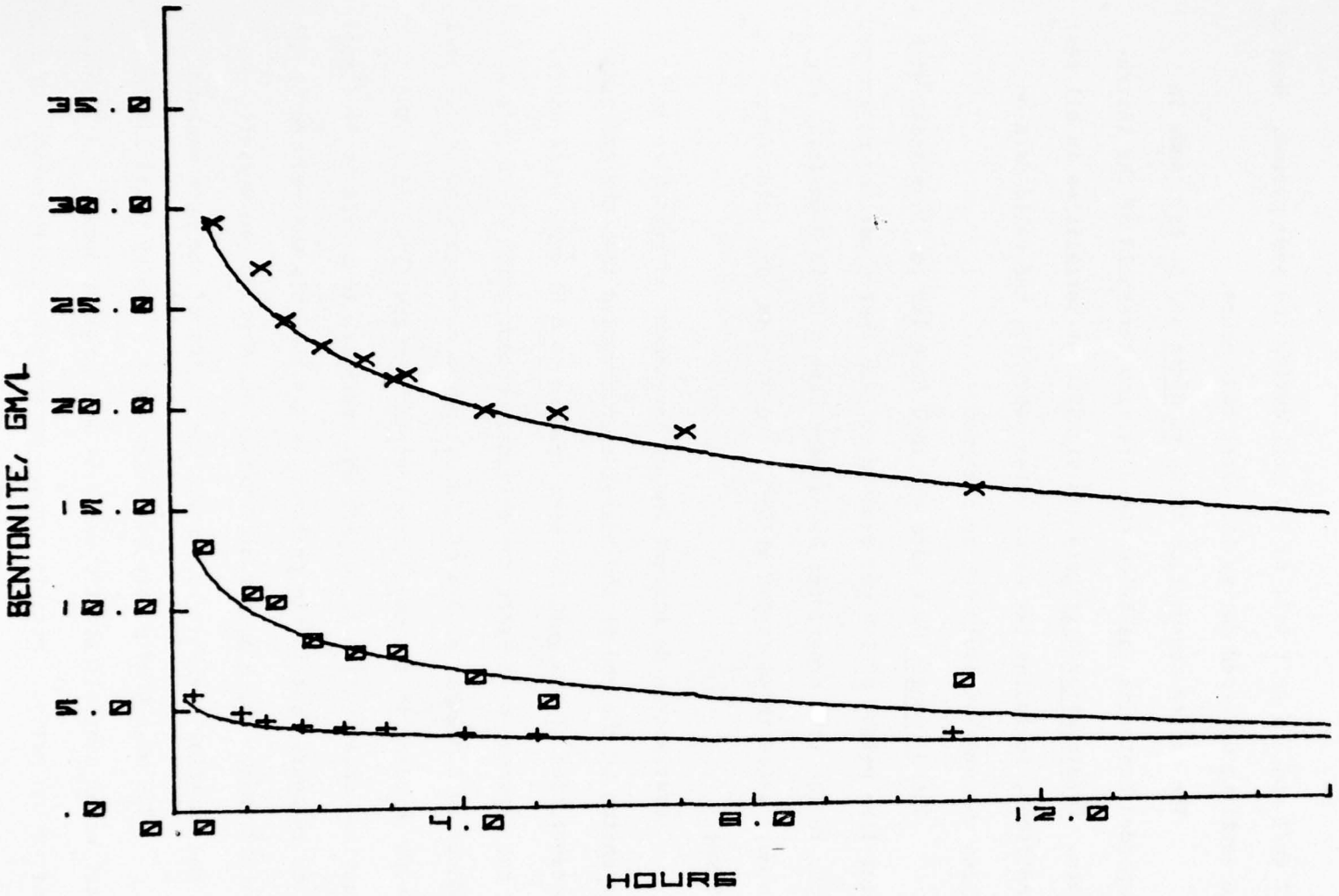

Fig. 33. Settling rate of suspended bentonite in 3-liter test containers in absence of animals.

3.2 gm/l and 5.5 gm/l fell to 2.7 gm/l during the same period. Most of the settling occurred during the first four hours.

The oxygen consumption of N. succinea and S. laticauda in suspended bentonite was below the detection threshold of the instruments. Cymatogaster aggregata had significant mortalities in all test conditions, including the clear water controls, and valid data on oxygen consumption could not be obtained.

The M. edulis data were obtained from 110 to 130 mussels with a total live weight of 100 gm, exposed to each test condition as groups. Those in the 11°C experiment had a mean length of 18.8 mm (std. dev. 2.6 mm), while those tested at 18°C were 22.5 mm long (std. dev. 2.5 mm).

Gravimetric determinations of the amount of bentonite in suspension at the end of the mussel test revealed that the starting concentration of 5.5 gm/l had been reduced to 0.01 gm/l in 14 hours. In the presence of mussels the original concentration of 13 gm/l was reduced to 2.0 gm/l, or 39% of the estimated concentration of 3.2 gm/l due to settling in the empty respiration chambers (Fig. 33). The starting concentration of 30 gm/l was estimated to settle to 14.4 gm/l after 14 hours, but in the presence of mussels this was reduced by 12% to 12.7 gm/l. While some sedimentation occurred on the mussels, most of the material accumulated beneath the bottom of the mesh baskets.

The mean hourly rate of oxygen consumption by M. edulis in the clear water controls at 11°C was 1.52 mgm O_2/gm/hr during the 12-hour observation period. Mussels exposed to starting concentrations of 5.5 gm/l, 13 gm/l and 30 gm/l consumed oxygen at mean rates of 0.91 mgm

O_2/gm/hr, 0.69 mgm O_2/gm/hr and 0.23 mgm O_2/gm/hr, respectively (Appendix E, Figs. E1-E4). Table XXII, which contains the statistics from the regression analyses illustrated in Figs. E1-E4, shows that the 95% confidence limits on the regression coefficients at 0, 5.5 and 30 gm/l do not overlap, indicating different rates of oxygen consumption. Figure 34 compares the regression estimates for cumulative oxygen consumption over time in the four suspended bentonite concentrations at 11°C. Except at 30 gm/l all showed an increasing rate of consumption over time, with the most rapid increase at 5.5 gm/l.

Table XXIII contains the statistics from the regression analyses of cumulative oxygen consumption over time by M. edulis in four suspended bentonite concentrations at 18°C. Figure 35 compares the regression estimates from these analyses, which are illustrated in detail in Appendix E, Figs. E5-E8. The oxygen consumption of mussels exposed to an original suspended bentonite concentration of 5.5 gm/l was not different from that of the controls. The mean rate of consumption was 2.07 mgm O_2/gm/hr in the clear water controls, 2.05 mgm O_2/gm/hr in 5.5 gm/l suspended bentonite, 1.27 mgm O_2/gm/hr or 61% of the controls at 13 gm/l and 0.64 mgm O_2/gm/hr or 31% of the controls at 30 gm/l suspended bentonite. At this concentration one mussel died in each of two replicates, neither of which were included in the data analysis. In contrast to the results at 11°C, the rates of consumption were constant in all test conditions at 18°C.

The results of the factorial analyses of variance considering the effects of suspended bentonite concentration, temperature and exposure time on the oxygen consumption of M. edulis are presented in

Table XXII

Relevant statistics from regression analyses of cumulative oxygen consumption by Mytilus edulis at 11°C during exposure to three levels of suspended bentonite and to clear seawater (controls).

Treatment	r^2	y-intercept	Regression coefficient	95% conf. limits on regression coefficient	df	F for linear regression
controls	.977+	- .652	1.365	1.184-1.545	1,8	***
5.5 gm/l	.973+	-2.231	1.899	1.639-2.159	1,8	***
13 gm/l	.973+	-1.924	1.556	1.280-1.831	1,8	***
30 gm/l	.955++	- .418	.279	.208- .349	1.8	***

+ Indicates best fitting line obtained by ln-ln regression.

++ Indicates best fitting line obtained by arithmetic regression.

*** $p<.001$

Table XXIII

Relevant statistics from regression analyses of cumulative oxygen consumption by Mytilus edulis at 18°C during exposure to three levels of suspended bentonite and to clear seawater (controls).

Treatment	r^2	y-intercept	Regression coefficient	95% conf. limits on regression coefficient	df	F for linear regression
controls	.998++	-1.061	2.115	2.015-2.215	1,10	***
5.5 gm/l	.997++	-1.988	2.225	2.098-2.351	1,10	***
13 gm/l	.990++	-1.891	1.335	1.199-1.470	1,10	***
30 gm/l	.979++	-1.579	.7347	.628- .841	1,10	***

++ Indicates best fitting line obtained by arithmetic regression.

*** $p<.001$

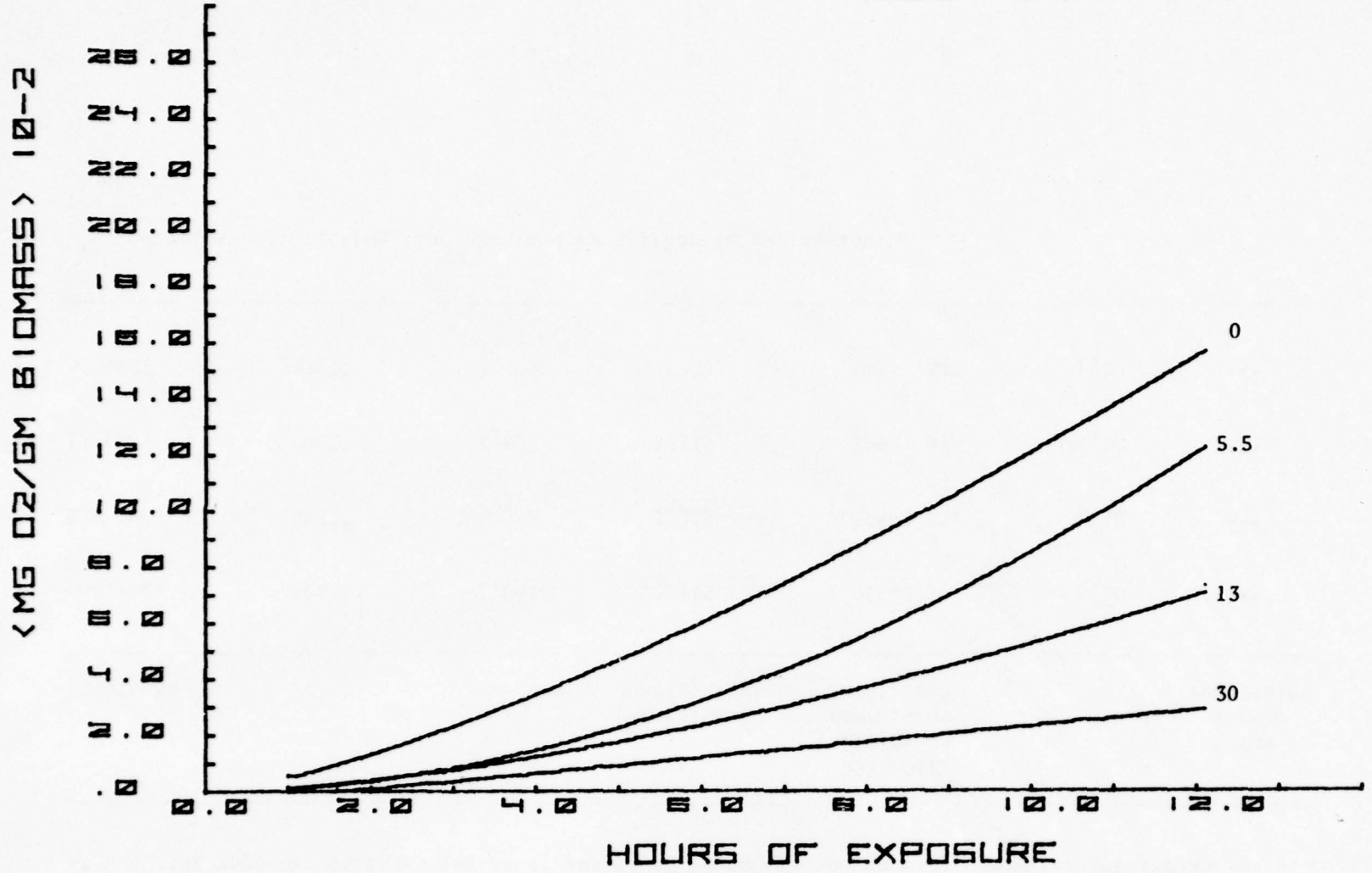

Fig. 34. Cumulative oxygen consumption of 19 mm Mytilus edulis during exposure to four concentrations of suspended bentonite at 11°C and 29% salinity. Numbers on the lines indicate suspended bentonite concentrations in gm/l at the start of the experiment.

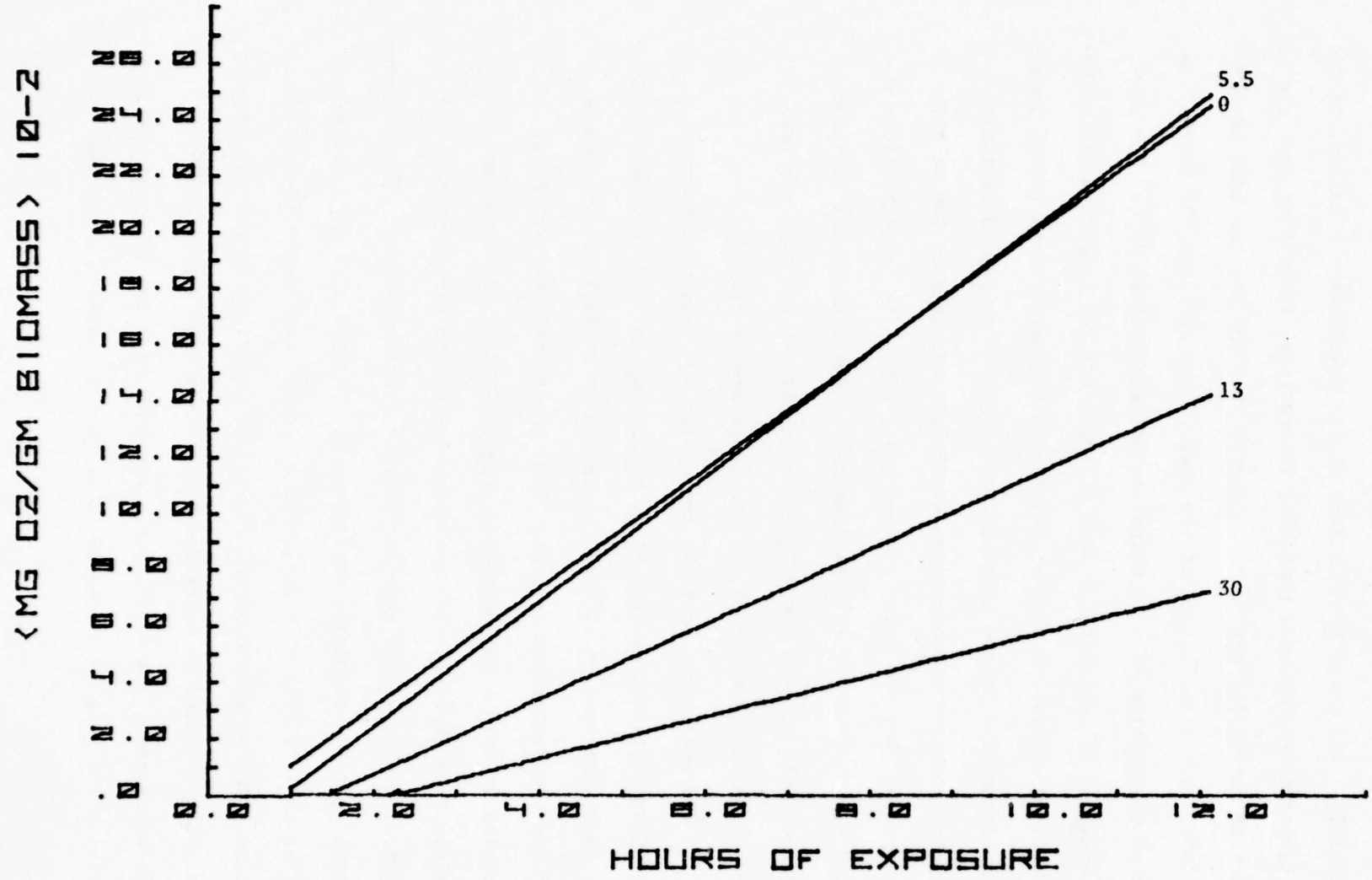

Fig. 35. Cumulative oxygen consumption of 22 mm Mytilus edulis during exposure to four concentrations of suspended bentonite at 18° and 29‰ salinity. Numbers on the lines indicate suspended bentonite concentrations in gm/l at the start of the experiment.

Table XXIV. All three factors gave highly significant F values, as did the temperature-suspended bentonite interaction, indicating that the effect of increasing bentonite concentrations was not the same at 11°C and 18°C. The significance of the time factor reflects the increasing rate of consumption at 11°C which was not apparent at 18°C. The one-way analysis of variance and mean contrast for the suspended bentonite-temperature conditions (Table XXV) showed a general trend toward lower oxygen conumption at low temperature and higher suspended bentonite concentrations. The consumption at 30 gm/l was equally low at both temperatures, but for all other suspended bentonite concentrations was significantly lower at 11°C than at 18°C. The analysis of variance confirmed that at 18°C the oxygen consumption at 5.5 gm/l was not different from the controls, as was indicated by Fig. 35.

The statistics from the regression analyses of cumulative oxygen consumption over time by C. nigricauda in four suspended bentonite concentrations at 18°C are contained in Table XXVI. Figure 36 compares the regression estimates from these analyses, which are presented in detail in Appendix E, Figs. E9-E12. The data were obtained from three to five non-ovigerous shrimp with a mean live weight of 0.9 gm tested together in each reaction vessel. The 95% confidence interval about the regression coefficient at 0 gm/l did not overlap the interval at 1 gm/l, but all other confidence intervals did overlap. The magnitude of the regression coefficients did not seem to be directly related to suspended bentonite concentration, and the differences among the coefficients may have been due to experimental variation. Figure 36 indicated no pattern of changes in oxygen

Table XXIV

Factorial analysis of variance table for the oxygen consumption experiment with Mytilus edulis. Observations were mg O_2 consumed per gm live weight during four three-hour time intervals.

Source	df	SS	MS	F
Suspended bentonite concentration	3	532	117	50.6**
Temperature	1	227	227	64.8**
Time	3	203	68	19.3**
Suspended bentonite-temperature interaction	3	39	13	3.7*
Suspended bentonite-time interaction	9	29	3	1 ns
Temperature-time interaction	3	16	5	1.5 ns
Suspended bentonite-temperature-time interaction	9	29	3	1 ns
Error	192	673	3	

ns: not significant p .05

*: p<.05

**: p<.01

Table XXV

One-way analysis of variance table and mean contrast for the comparison of oxygen consumption of Mytilus edulis under the 8 interdependent combinations of suspended bentonite concentration and temperature. In the mean contrast those means underlined by the same line are not different at the .05 probability level, while those not joined by the same line are significantly different.

Anova Table

Source	df	SS	MS	F
Suspended bentonite-temperature conditions	7	798	114	25.9**
Error	216	950	4	

** p<.01

Mean Contrast

Suspended bentonite-gm/l	30	30	13	5.5	13	0	5.5	0
Temperature-^{o}C	11	18	11	11	18	11	18	18
Mean Oxygen consumption-mg O_2/gm live weight	0.69	1.93	2.06	2.72	3.81	4.56	6.14	6.21

Table XXVI

Relevant statistics from regression analyses of cumulative oxygen consumption by Crangon nigricauda at 18°C during exposure to three levels of suspended bentonite and to clear seawater (controls).

Treatment	r^2	y-intercept	Regression coefficient	95% conf. limits on regression coefficient	df	F for linear regression
controls	.997+	-1.125	1.383	1.270-1.495	1,6	***
1 gm/l	.999+	- .989	1.201	1.141-1.262	1,6	***
3 gm/l	.996+	- .775	1.283	1.168-1.399	1,6	***
10 gm/l	.996+	- .925	1.204	1.099-1.309	1,6	***

+ Indicates best fitting line obtained by ln-ln regression.

*** $p<.001$

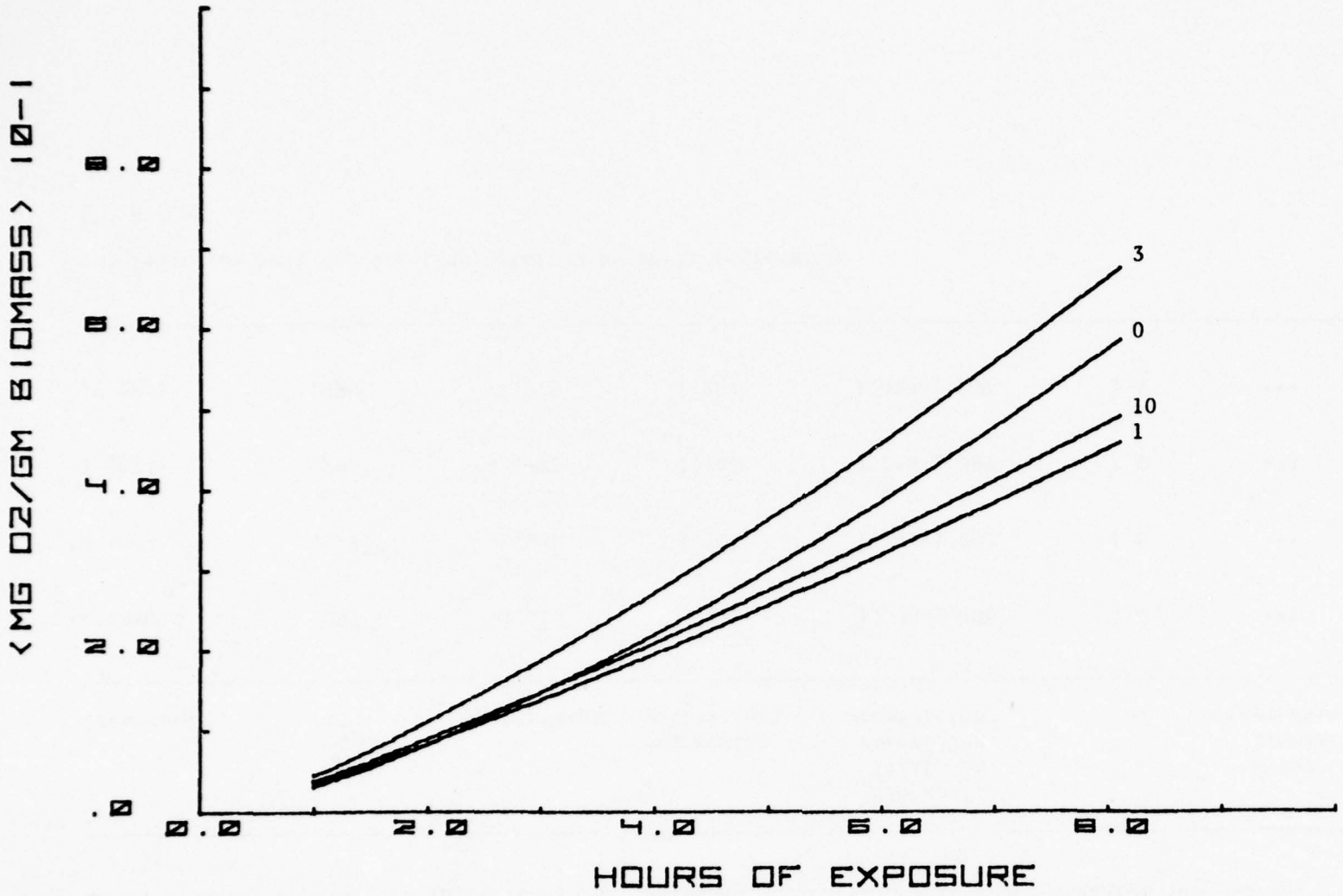

Fig. 36. Cumulative oxygen consumption of 4 cm Crangon nigricauda during exposure to four levels of suspended bentonite at 18°C and 28‰ salinity. Numbers on the lines indicate suspended bentonite concentrations in gm/l at the start of the experiment.

consumption with increasing suspended bentonite concentration and the analysis of variance showed the effects of suspended bentonite to be not significant (Table XXVII). The influence of exposure time significantly affected oxygen consumption and the mean contrast revealed higher values during the later time intervals, indicating an increasing rate of consumption.

The statistics in Table XXVIII are from the regression analyses of the cumulative oxygen consumption over time of M. saxatilis in four suspended bentonite concentrations at 18°C, as illustrated in Appendix E, Figs. E13-E16. The regression estimates from these analyses are compared in Fig. 37. Observations were on the response of individual fish weighing about 7.3 gm. There were no statistical differences among the regression coefficients at 0 gm/l, 2 gm/l and 4 gm/l, and the regression estimates all appear similar in Fig. 37. However, the regression coefficient at 10 gm/l was significantly different from the others, indicating a more rapid rate of oxygen consumption. One death occurred in the highest suspended bentonite concentration.

The factorial analysis of variance (Table XXIX) showed significant influences on oxygen consumption due to the interaction of suspended bentonite concentration and exposure time. The one-way analysis of variance and mean contrast (Table XXX) indicated that oxygen consumption was highest in the higher suspended solids concentrations, especially after longer exposures.

DISCUSSION

Few deaths were observed early in any of the suspended bentonite

Table XXVII

Factorial analysis of variance and mean contrast for the oxygen consumption experiment with Crangon nigricauda. Observations were mg O_2 consumed per gm live weight during four 2-hour time intervals. The mean contrast was by Tukey's W-procedure at the .05 level.

Anova Table

Source	df	SS	MS	F
Suspended bentonite concentration	3	4	1	1.7 ns
Time	3	22	7	9.9**
Suspended bentonite-time interaction	9	13	1	1.9 ns
Error	112	82	1	

ns: not significant p .05

**: p<.01

Mean Contrast

3-4 hours	1-2 hours	7-8 hours	5-6 hours
1.06	1.13	1.59	2.09

Table XXVIII

Relevant statistics from regression analyses of cumulative oxygen consumption by Morone saxatilis at 18°C during exposure to three levels of suspended bentonite and to clear seawater (controls).

Treatment	r^2	y-intercept	Regression coefficient	95% conf. limits on regression coefficient	df	F for linear regression
controls	.986++	-1.956	3.628	3.005-4.251	1,6	***
2 gm/l	.980++	.580	3.524	2.811-4.237	1,6	***
4 gm/l	.979++	-1.210	3.653	2.899-4.407	1,6	***
10 gm/l	.996++	-3.134	5.361	4.896-5.826	1,6	***

++ Indicates best fitting line obtained by arithmetic regression.

*** $p<.001$

Table XXIX

Factorial analysis of variance table for the oxygen consumption of Morone saxatilis at 18°C. Observations were mg O_2 consumed per gm live weight during each of two consecutive two-hour time intervals.

Anova Table

Source	df	SS	MS	F
Suspended bentonite concentration	3	50	17	5.8*
Time	1	20	20	6.9*
Suspended bentonite-time interaction	3	99	33	11.4**
Error	40	116	3	

* $p<.05$

** $p<.01$

Table XXX

One-way analysis of variance table and mean contrast for the comparison of oxygen consumption of Morone saxatilis under the 8 interdependent combinations of suspended bentonite concentration and exposure time. In the mean contrast those means underlined by the same line are not different at the .05 probability level, while those not joined by the same line are significantly different.

Anova Table

Source	df	SS	MS	F
Suspended bentonite-time conditions	7	169	24	8.4**
Error	40	116	3	

** $p<.01$

Mean Contrast

Suspended bentonite-gm/l	4	2	0	0	10	4	2	10
Time interval-hours	1-2	3-4	1-2	3-4	1-2	3-4	1-2	3-4
Mean oxygen consumption-mg O_2/gm live wt	4.5	5.2	5.7	7.2	7.5	8.6	8.6	11.0

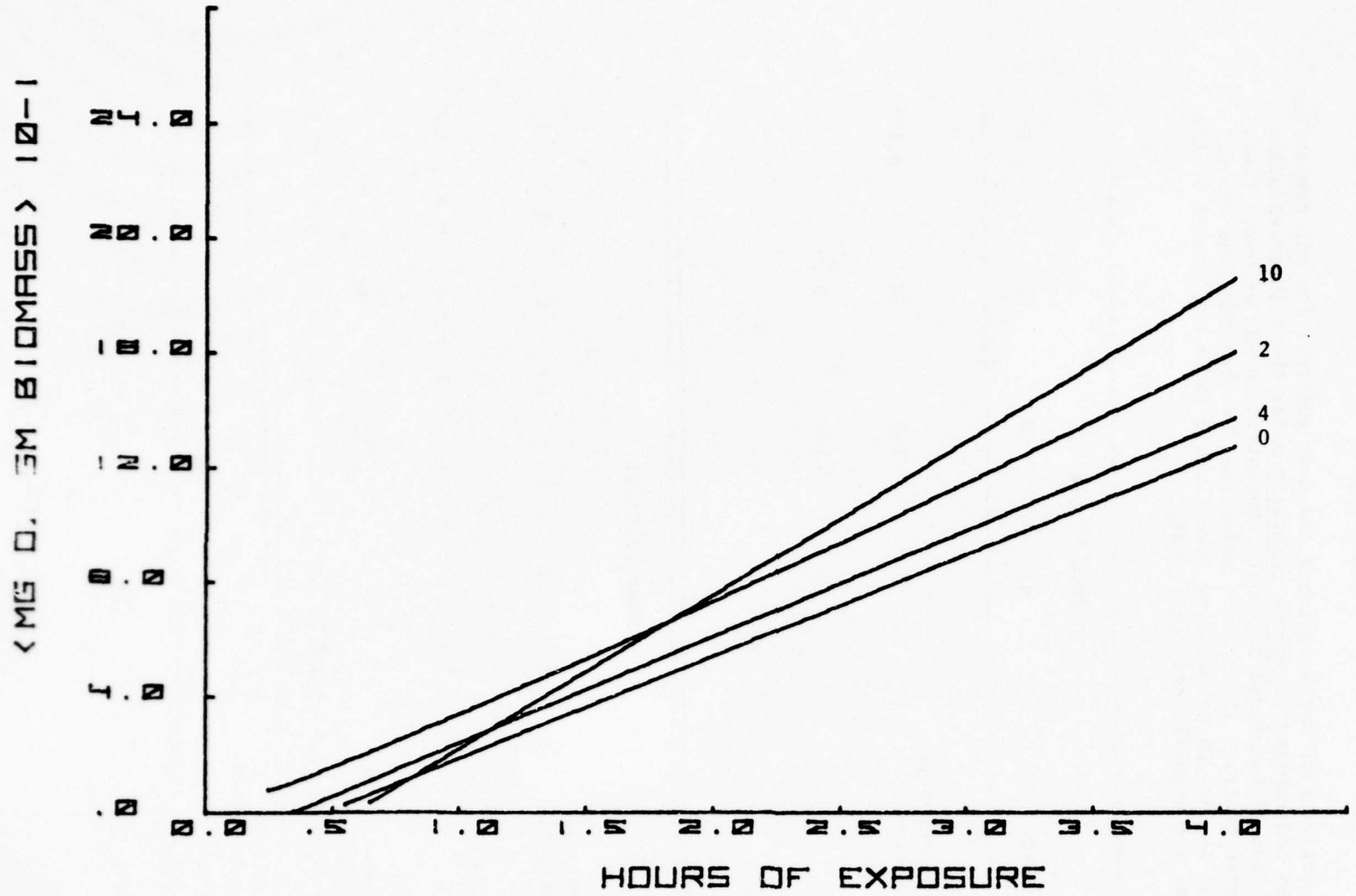

Fig. 37. Cumulative oxygen consumption of 6 cm Morone saxatilis during exposure to four concentrations of suspended bentonite at 18°C and 24% salinity. Numbers on the lines indicate suspended bentonite concentrations in gm/l at the start of the experiment.

mortality experiments with M. edulis. Mortalities usually reached 10% only after approximately 100 hours of exposure, but then occurred rapidly, resulting in sharply declining LC_x estimates. This delayed but rapid mortality may indicate that the mussels remained closed during the first few days of the test, dying only when physiological needs forced them to open, exposing themselves fully to the adverse experimental conditions. The much greater mortality observed at high temperatures than at low may result from a higher metabolic rate forcing the mussels to open sooner, thus hastening exposure to the bentonite particles. Another possibility is that M. edulis, being an estuarine filter feeder, has some inherent ability to deal with suspended solids, and died only when this capability was overwhelmed by high particle concentrations or long exposures. If so, the increased metabolic rate at higher temperatures would contribute to a more rapid breakdown of this ability.

In the oxygen consumption experiments it was assumed that the animals in the clear water controls were using the optimum amount of oxygen under the experimental conditions. Therefore a change from this level would indicate an effect of suspended solids on consumption. The highest oxygen consumption by M. edulis occurred in the controls at both temperatures, and decreased with increasing suspended bentonite concentration. This may indicate coating or irritation of the gill surface by the particles or a progressive decrease in pumping activity. That the mussels did not all cease activity and close the shells completely was indicated by the fact that some oxygen consumption was measured under all experimental conditions. Active filtration was also

demonstrated by the removal of all particles from suspension in the starting concentration of 5.5 gm/l at 18°C. This was in agreement with Fox, Sverdrup and Cunningham (1937), who showed that M. californianus, a closely related species, opened almost immediately and began filtering sediment from suspension when placed in concentrations of 4 gm/l. Davids (1964) found that if M. edulis were moved from a high concentration of algal cells to a lower one the pumping rate increased, and decreased if a suspension was followed by a more concentrated one. Thus the decrease in oxygen consumption we observed with increasing suspended bentonite concentration may be due to an inhibition of pumping activity.

Although determinations of mechanisms of effect was beyond the scope of this research, the data indicated that the oxygen uptake of M. edulis decreased with increasing suspended bentonite concentration. Such an interference, which is suggested as the primary impact of suspended solids, would have been exaggerated by increased respiration rates at higher temperatures, producing the greater mortality observed at 18°C than at 10°C. The continuation of deaths through the post-exposure periods after all experiments suggests that deaths were related to some sort of long-term damage.

The experimental conditions resulted in loss of byssal attachments by M. edulis after exposure times much shorter than those causing death (Tables X and XIX). Reish and Ayers (1968) found that the number of byssal threads produced by M. edulis decreased greatly at low dissolved oxygen levels, while mussel survival remained high. It is perhaps significant that a phenomenon Reish and Ayers attributed to

physiological stress resulting from low dissolved oxygen was found to be associated with increased suspended solids levels in the present experiments. The loss of byssal attachments may be an early and sensitive indicator of effective death. Mussels which became detached in the field and fell to the bottom would perhaps be more susceptible to predation and certainly more subject to covering by sedimentation. This would be especially likely near a dredging operation which created suspended solids high enough to cause detachment initially.

Our studies showed that the suspended solids tolerance of C. nigricauda was markedly influenced by temperature, even though survival in the clear water control tanks was little affected. Haefner (1969) has shown that C. septemspinosa, a related Atlantic Coast species, also suffered little mortality in clear, oxygen saturated water of the temperature and salinity used in our experiments. However, under these conditions 2-3 ppm dissolved oxygen increased mortality of non-ovigerous C. septemspinosa to 20% to 30% and of ovigerous females to 40% to 50% (Haefner, 1970). Our experiments indicated that C. nigricauda may not be quite so sensitive to low dissolved oxygen, since 13% mortality occurred among all shrimp exposed to 5 ppm dissolved oxygen in the clear water control tanks and 19% of the control animals died at 2 ppm dissolved oxygen. However, the influence of dissolved oxygen on the suspended solids tolerance of C. nigricauda was very great, as can be seen by comparing the results at saturation (Fig. 16), 5 ppm (Fig. 21), and 2 ppm dissolved oxygen (Fig. 22).

There was no clear indication of any effect of suspended solids

on the oxygen consumption of C. nigricauda. A longer exposure period might have shown some effect since these shrimp may be able to cope with suspended solids for a short time by some mechanism which is overcome by longer exposure to high concentrations. Such a compensating mechanism is possessed by the mysid Gnathophausia ingens which increases its oxygen uptake capability as dissolved oxygen in the water decreases, thus maintaining its characteristic rate of oxygen consumption (Childress, 1968).

The mortality experiments with S. laticauda showed a high tolerance to suspended solids. The multifactor analysis of variance indicated that neither temperature, dissolved oxygen nor suspended solids concentration had a significant effect on survival. The few deaths which did occur were apparently random and not related to the experimental variables.

Mortalities of N. succinea were significant in the clear water controls, and although higher in the experimental aquaria, bore no relationship to increasing suspended solids concentration. The absence of a substratum into which they could burrow left the polychaetes exposed in the test containers. The secreted mucous coated the mesh of the containers, causing sedimentation and covering the worms with compacted bentonite, in which they failed to establish tubes or burrows. Although there were indications of somewhat higher mortality in the experimental aquaria than in the controls, the presence of significant unidentified stresses other than the experimental variables was obvious. No definite conclusions concerning the tolerance of N. succinea to suspended solids could be drawn.

Both species of fish were more sensitive to suspended solids than any of the invertebrates included in the bentonite experiments. The fish survived longer at high temperatures than at low, in contrast to all the invertebrates. The experiments involving temperature were conducted at a time when collection temperature nearly equalled the high test temperature and acclimation was required only for the cold tests, perhaps stressing these fish. This seems unlikely since the acclimation was done slowly, no fish died during the process and mortalities in the cold control aquaria were negligible. One alternative explanation would be the presence of some compensating mechanism which was more effective at the higher metabolic rates associated with high temperatures. Decreases in the sensitivity of fish to certain chemical toxins at higher temperatures have been discussed by Black et al. (1973).

Cymatogaster aggregata, the most sensitive species studied, was affected by increasing suspended solids concentrations under all temperature and dissolved oxygen conditions tested. The great stress from low dissolved oxygen partly masked the effects of temperature and suspended solids, but at higher dissolved oxygen levels survival was primarily dependent on these factors. In the multifactor experiment the mean survival time of C. aggregata was significantly reduced under several combinations of conditions which might conceivably occur on a dredging site (Table XVII). For example, the mean survival time was only 15 hours in 2.2 gm/l at 18°C and 2 ppm dissolved oxygen.

The study of oxygen consumption by M. saxatilis, conducted with suspended bentonite concentrations similar to the mortality experiments,

revealed increased consumption at the highest suspended solids level. Whether this was a long-term phenomenon or the result of a temporary increase in oxygen requirements or uptake capability could not be determined. Sherk et al. (1974), who tested larger M. saxatilis under different experimental conditions at constant swimming speeds, found that oxygen consumption decreased with relatively small increases in suspended solids. Although both studies showed an effect of suspended solids on oxygen consumption, the differences in results indicate the need for more research. Sherk et al. (1974), who also studied hematology and gill morphology, felt that suspended solids produced mortality in fish by several mechanisms, although the ultimate cause of death was anoxia in all cases. A coating of fine particles would remove the respiratory epithelium from contact with the water, producing a locally anoxic condition at the gill surface resulting in asphyxiation. Larger particles may be trapped by the gill lamellae, and reduce water circulation at the primary site of gas exchange. Suspended solids could also limit effective gas exchange by mechanical injury to the gill surfaces.

Although the mechanism by which the fish were killed in the present experiments was not investigated, major structural damage probably did not occur. Both species had nearly total survival during all the post-exposure periods, indicating either the absence of serious physical damage or the presence of an efficient repair mechanism. The hypothesis of Sherk et al. (1974) concerning gill coating and/or clogging seems a more likely explanation of death. In a hematological study of fish under respiratory stress, Hall et al. (1926) suggested

that death by asphyxiation might be related to the concentration of toxic byproducts in the blood through a loss of fluid to the tissues.

In none of the experiments with any species could the magnitude of the LC_{50} value be predicted from the LC_{20} or LC_{10} values, or vice versa. Sherk et al. (1974) studied a variety of estuarine fish and found that the range of concentration between the LC_{10} and LC_{90} values did not necessarily indicate the magnitude of the LC_{50} value. These results indicate the necessity for studying the tolerance of the most sensitive members of a population.

There seemed to be some correlation between normal habitat of the species studied and sensitivity to suspended solids, although no phylogenetic correlations were apparent. No species which lives primarily in close association with mud bottoms was found to be sensitive. While many species occupying other habitats were also highly tolerant, all species shown to be sensitive to high suspended solids were either invertebrates occurring predominantly on sandy bottoms or in fouling communities, or fish not intimately associated with the bottom. Such species are probably subjected to high suspended solids less often than those living in or on mud bottoms, and may have less well developed mechanisms for dealing with such occurrences. It should be stressed that this research did not emphasize sublethal stresses and that lack of mortality in 10 days does not imply the absence of significant effects.

Our results indicate that high suspended solids concentrations would be less harmful in winter than in summer. The lower temperature would increase the solubility of oxygen in the water, minimize any sag

related to oxygen demand of resuspended sediments, and lower the oxygen requirements of the animals, all contributing to survival ability. The tolerance of both the fish and invertebrates to suspended solids was greater at high dissolved oxygen levels, while low temperature increased the tolerance of the invertebrates but decreased that of the fish. However, the differences in magnitude between the effects of temperature and dissolved oxygen indicate that the negative effect of low temperature would be offset by the increased tolerance at high dissolved oxygen levels. In winter there would also be fewer actively reproducing adults and fewer larvae and immature stages present, which may be more sensitive to lower suspended solids concentrations than adults (Davis, 1960; Davis and Hidu, 1969; Morgan et al., 1973; Schubel and Wang, 1973).

These experiments were conducted in a laboratory environment which obviously differed from estuarine conditions. However, the use of an open system allowing relatively undisturbed long-term testing under carefully monitored physical conditions, coupled with frequent biological observations permitting determination of rates of effect, overcome many major concerns about the type of research. "It cannot be said how far the results of this laboratory study are directly applicable to the survival of animals in waters polluted with inert solid material, but a stress which reduces survival chances in one environment will probably reduce them to some extent in another, although not necessarily to the same degree" (Herbert and Merkins, 1961).

BIOLOGICAL EFFECTS OF RAPID SEDIMENT DEPOSITION

Studies of the ability to survive rapid sediment overlayering resulting from dredging operations were a secondary part of the project. The species used in the suspended solids study were also the subject of burial research, whose objective was the determination of short-term survival ability when covered by various depths of material.

HISTORICAL BACKGROUND

Much of the literature on animal responses to burial is largely anecdotal and not directed toward specific questions. Many of the controlled studies of burial and escape behavior deal with commercially important bivalves. Glude (1954) conducted field burial experiments with Mya arenaria 9 to 50 mm long covered by up to 22 cm of a variety of sediments. He found the probability of survival to vary inversely with depth of burial and directly with size of the organism. Survival was lowest in silt, higher in sand and highest in silty sand, and was higher in winter than in summer.

Schafer (1962), who described the exhuming behavior of many marine species, including polychaetes, actiniarians, scaphopods, gastropods and bivalves, stated that M. arenaria could escape from 10 cm of sand in 2 to 10 hours. He showed that surface dwelling bivalves, such as mussels and scallops, could not cope with large amounts of sediment and were not found in areas of sedimentation. Mussels were able to elevate themselves slightly via the byssus, overcoming a few millimeters of sedimentation, and scallops could eject

some sediment from the mantle cavity by valve movement.

In an X-radiography study Shulenberger (1970) concluded that Gemma gemma, a small bivalve, could cope with coverings up to 23 cm with sand and 5.7 cm with silt. Survival for up to 6 days was possible under a variety of burial conditions.

Kranz (1974) conducted laboratory and field studies of 25 species of bivalves to determine the effects of catastrophic burial and its palaecological significance. He demonstrated that the exhuming ability of bivalves is closely related to their life habit. Borers, deep burrowing adult siphonate suspension feeders and suspension feeding epifaunal forms were generally unable to escape sediment coverings thicker than 1 cm. However, shallow burrowing siphonate suspension feeders and young deep burrowers were usually able to escape from under 10 cm to 50 cm of their native sediment. A radical change from the native sediment type could be highly lethal by reducing the effective burrowing ability, and often burial in only 1 cm of an exotic sediment was fatal. Kranz found no simple correlation between living depth and exhuming ability. Temperature, salinity and oxygen concentration seemed to have little effect on exhuming ability except near the extremes of a species tolerance. The nature of the native and deposited sediments and the life habits of the bivalves in question were the most important factors in determining survival.

The damage to market-sized oysters Crassostrea virginica by sedimentation from a dredging operation was studied by Rose (1973). He found the oysters suffered 57% mortality within 595 m of the spoil site, where they were covered with 2-15 cm of sediment. This compared to 17%

mortality during the same period in the remainder of the oyster bed where little sedimentation occurred. The observed mortality of 40% compared to a calculated theoretical mortality of 48%, which was estimated to have been produced by sedimentation resulting from dredging if other mortality-inducing factors had not been operative. The information presented in this paper was the subject of litigation in Louisiana.

Few observations are available on the responses of soft-bodied organisms to either natural or experimental burial. Exceptions include studies of the onuphid polychaetes Diopatra cuprea (Myers, 1972), and Nothria elegans (Oliver and Slattery, 1973), both of which were capable of burrowing upward through an accumulation of 30 cm of sediment. Similarly, the terebellid polychaete Pista pacifica is capable of extending its tube up through at least 25 cm of sediment (personal communication, Thomas Ronan).

Oliver and Slattery (1973), who conducted burial experiments with native and exotic sediments on a subtidal community, reported that all small pelecypods and crustaceans, such as cumaceans and harpacticoid copepods, were killed by deposition of 15 cm of sediment. Large bivalves, such as Tresus nuttallii, established "blowholes" to the surface of the sediment. When compared with a nearby reference area the number of individuals was reduced 50% by the sediment deposition. All organisms that survived the sedimentation were normal residents of, or actively burrowed into, the lower sediment strata. The overall impact on the community was generally least under depositions of the native sediment.

METHODS AND MATERIALS

The burial experiments were conducted in 45 liter wood-and-glass aquaria provided with sandy substrata in which the animals were allowed to establish themselves before the experiment began. The aquaria had a once-through flow of water with a one-hour turnover time. Salinity was controlled by a pair of valves on the salt and fresh water head tanks, as described for the suspended solids facility.

The experimental material was the same bentonite used in the suspended solids research. This was mixed with seawater to a 50% slurry on a weight per volume basis. A pilot study was conducted to determine how much of this slurry had to be added to the aquaria to settle to the desired thickness.

The animals used in the burial experiments were taken from the collections made for the three-factor suspended solids experiment, and had no previous laboratory exposure to suspended or deposited bentonite. The two species of fish were tested simultaneously in the same aquaria and all the invertebrates were tested together.

Before beginning an experiment 10 cm of fine to medium sand, grain size 150μ to 350μ, with approximately 10% silt and clay content and containing no macrofauna was placed in the aquaria. Salinity and flow rate of the water were adjusted, and animals were then counted into the aquaria and allowed 24 hours to establish themselves in the new environment. The experiment was begun with the rapid pouring of enough slurry into the aquaria to produce deposits of 2, 4, 6 and 8 cm thickness. One aquarium, to which no slurry was added, was maintained

as a control. The behavior of the animals was observed immediately after slurry introduction, periodically during the next few hours and then daily. When the experiments were terminated after 96 hours (4 days), the large animals were individually removed, then the material in the aquaria was passed through a 1 mm screen to retain the smaller organisms. The animals were sorted by species and the numbers alive and dead were recorded.

RESULTS AND DISCUSSION

Data on the conditions under which the animals were collected, held in the laboratory and tested are presented in Appendix F, Tables FI and FII. The results of the experiment are presented in Table XXXI, showing the number of animals tested, the number of deaths and the percent dead 96 hours after each depth bentonite had been deposited.

No fish of either species died during the experiment. When the bentonite slurry was added all fish were immediately coated with mud, perhaps adhering to mucous on the body surfaces. This had disappeared within 1/2 to 1 hour as the water cleared and the mucous was sloughed off. Throughout the remainder of the test the appearance and behavior of the fish seemed normal.

The C. nigricauda, which had burrowed normally into the substratum in all aquaria, became active as soon as the slurry was introduced and swam continuously until the bentonite had compacted enough to provide some support. However, the shrimp in the test aquaria were never able to rest without sinking slowly into the mud,

Table XXXI.

Results of the deposition experiments, showing the species and the number tested, number dead and percent dead 4 days after the bentonite was deposited to the specified depths.

Species	0 cm orig. no.	0 cm no. dead	0 cm o/o dead	2 cm orig. no.	2 cm no. dead	2 cm o/o dead	4 cm orig. no.	4 cm no. dead	4 cm o/o dead	6 cm orig. no.	6 cm no. dead	6 cm o/o dead	8 cm orig. no.	8 cm no. dead	8 cm o/o dead
Morone saxatilis 5.5-8 cm	20	0	0	20	0	0	20	0	0	20	0	0	20	0	0
Cymatogaster aggregata 5.5-7.5 cm	10	0	0	10	0	0	10	0	0	10	0	0	10	0	0
Crangon nigricauda 3-5 cm	10	0	0	10	0	0	10	0	0	10	0	0	10	0	0
Synidotea laticauda "adult"	10	0	0	10	0	0	10	1	10	10	2	20	10	2	20
Mytilus edulis 2-3 cm	10	0	0	10	0	0	10	1	10	10	6	60	10	6	60

and moved about more than those in the controls. No C. nigricauda died during the experiment (Table XXXI).

The isopods S. laticauda behaved similarly to the shrimp, swimming until the bentonite was compacted sufficiently for them to crawl about on it. One of the ten isopods was not able to rise through the 4 cm of slurry as it was added to the aquarium, and was killed. Twenty percent mortality occurred when 6 cm and 8 cm of bentonite slurry were added (Table XXXI).

All M. edulis had firm byssal attachments when the bentonite was deposited on them. However, at the end of the experiment the mussels covered with 2 cm of bentonite had few byssal attachments and no surviving mussels buried deeper than 2 cm had any byssal threads. Those in the control aquarium remained firmly attached throughout the experiment. No deaths occurred under 2 cm of bentonite, but 10% mortality occurred under 4 cm and 60% mortality was observed when 6 cm and 8 cm of slurry were deposited on the mussels. These results indicate that near dredging operations, where suspended solids might reach concentrations high enough to cause loss of byssal attachments, mussels which fell to the bottom and were covered with deposited sediment would probably be killed.

The only deaths occurred among species typical of fouling communities, which are not normally subjected to sedimentation. The mortality of the mussels was in agreement with the principles concerning behavior and feeding type set forth by Schafer (1962) and Kranz (1974). Although no mortalities were noted among the C. nigricauda, the prolonged constant swimming by these organisms not accustomed to

such activity may be significant. In situations where new material is added continuously or at short intervals, as with pipeline or hopper dredge disposal, the surface may not compact sufficiently to support the shrimp. If not, exhaustion could result unless they were able to move away from the disturbed area.

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APPENDIX A

Animal Collection and Holding Data and Experimental Conditions for the Suspended Kaolin Mortality Experiments

APPENDIX A. KAOLIN MORTALITY EXPERIMENTS COLLECTION AND HOLDING DATA

Table AI. List of species tested; date, method and site of collection; and the salinity and temperature from which the animals were collected. Also presented is the length of time each species was maintained in the laboratory before testing began, and the salinity and temperature at which each was held.

SPECIES	COLLECTION DATA					HOLDING DATA		
	date	method	site	salinity ppt	temp. °C	days held in lab	salinity ppt	temp. °C
Strongylocentrotus purpuratus	mid-April 1973	hand	Bodega Bay	30-32	10-12	8	30-32	10-12
Crangon franciscorum	early Feb. 1974	otter trawl	Central S.F. Bay	14-16	12-14	3	14-16	12
Pagurus hirsutiusculus	late March 1974	hand	Central S.F. Bay	18-20	12-14	9	18-20	12-14
Sphaeroma pentodon	late March 1974	hand	Central S.F. Bay	18-20	12-15	9	18-20	12-14
Nassarius obsoletus	early Feb. 1974	otter trawl	Central S.F. Bay	12-15	12-14	3	14-16	12
Tapes japonica	late Feb. 1974	hand	Central S.F. Bay	12-15	12-15	3	12-14	11-12
Mytilus edulis (2.5 cm)	early Feb. 1974	hand	Central S.F. Bay	12-15	13-15	3	14-16	12
Mytilus edulis (10 cm)	early May 1973	hand	Bodega Bay	30-32	10-12	5	30-32	9-10

Appendix A, Table AI, Collection and Holding data (continued)

SPECIES	COLLECTION DATA					HOLDING DATA		
	date	method	site	salinity ppt	temp. °C	days held in lab	salinity ppt	temp. °C
Molgula manhattensis	early April 1974	otter trawl	San Bruno Shoal area	12-15	14-16	6	18-20	12-14
Styela montereyensis	early April 1974	otter trawl	San Bruno Shoal area	12-15	14-16	6	18-20	12-14
Mytilus californianus	early May 1973	hand	Bodega Bay	30-32	10-12	5	30-32	9-10
Ascidia ceratodes	late May 1973	otter trawl	Bodega Bay	30-32	10-11	10	31-33	8-9
Crangon nigromaculata	early June 1973	otter trawl	Bodega Bay	30-32	10-12	5	30-32	10-11
Crangon nigricauda	late May 1974	otter trawl	San Bruno Shoal area	14-16	12-14	8	17-18	13-15
Palaemon macrodactylus	late Feb. 1974	dip net	Mare Island area	8-12	11-13	3	12-14	11-12
Cancer magister	early Aug. 1973	otter trawl	Bodega Bay	30-32	11-13	6	30-32	10-11
Anisogammarus confervicolus	late Feb. 1974	dip net	Mare Island area	8-12	11-13	3	12-14	11-12
Neanthes succinea	late Feb. 1974	hand	Mare Island area	8-12	11-13	3	12-14	11-12

SPECIES	COLLECTION DATA					HOLDING DATA		
	date	method	site	salinity ppt	temp. °C	days held in lab	salinity ppt	temp. °C
Parophrys vetulus	late May 1974	otter trawl	San Bruno Shoal area	14-16	12-14	5	17-18	13-15
Cymatogaster aggregata	mid-May 1973	otter trawl	Bodega Bay	30-32	9-11	12	30-32	8-9
Cymatogaster aggregata	mid-April 1974	otter trawl	San Bruno Shoal area	12-15	13-15	7	13-15	12-14

APPENDIX A. KAOLIN MORTALITY EXPERIMENTS
EXPERIMENTAL CONDITIONS

Table AII. Summary of species tested, duration of the experiments, the salinity and dissolved oxygen common to all aquaria, the number of test aquaria used, and the suspended solids concentration, temperature and pH in each aquarium. Values presented are means ±1 standard deviation. Species grouped together in the table were tested simultaneously in the same aquaria.

Species	Test Conc. No.	Suspended Kaolin gm/l	Temp. °C	pH
Strongylocentrotus purpuratus	1	control	11.1±0.3	7.9±0.1
	2	11 ±1.2	11.0±0.1	7.7±0.1
Test length: 216 hours Salinity: 32±1 ppt Dissolved oxygen: 8±1 ppm	3	13 ±1.2	10.9±0.2	7.6±0.1
	4	22 ±2.2	10.9±0.2	7.4±0.1
	5	34 ±2.7	10.9±0.2	7.2±0.1
	6	65 ±5.2	10.9±0.2	7.0±0.1
	7	115±11.3	10.9±0.2	6.6±0.2
Crangon franciscorum	1	control	11.9±0.2	7.9±0.1
	2	11 ±3.0	11.9±0.2	7.7±0.2
Test length: 120 hours Salinity: 15±1 ppt Dissolved oxygen: 8±1 ppm	3	34 ±4.0	11.9±0.2	7.2±0.2
	4	100±19.2	11.8±0.4	6.6±0.1
Pagurus hirsutiusculus	1	control	12.3±0.2	7.6±0.1
	2	11 ±1.3	12.2±0.1	7.4±0.1
Sphaeroma pentodon	3	15 ±1.8	12.2±0.2	7.2±0.1
Test length: 288 hours Salinity: 21.6±.9 ppt Dissolved oxygen: 8±1 ppm	4	26 ±2.0	12.3±0.1	7.1±0.1
	5	38 ±3.0	12.3±0.1	6.8±0.1

Appendix A, Table AII, Experimental conditions (continued) 2

Species	Test Conc. No.	Suspended Kaolin gm/l	Temp. °C	pH
Sphaeroma pentodon (cont.)	6	72 ±5.5	12.4±0.3	6.6±0.1
	7	101 ±8.5	12.4±0.1	6.3±0.1
Nassarius obsoletus	1	control	11.9±0.2	7.9±0.1
Mytilus edulis (2-5 cm)	2	11 ±3.0	11.9±0.2	7.7±0.2
Test length: 120 hours	3	34 ±4.0	11.9±0.2	7.2±0.2
Salinity: 15±1 ppt Dissolved oxygen: 8±1 ppm	4	100±19.2	11.8±0.4	6.6±0.1
Tapes japonica	1	control	11.5±1.4	7.7±0.2
Test length: 240 hours	2	9 ±0.9	11.1±1.4	7.5±0.2
Salinity: 13.2±.5 ppt Dissolved oxygen: 8±1 ppm	3	12 ±1.5	11.1±1.4	7.4±0.2
	4	31 ±2.7	11.2±1.3	7.1±0.1
	5	102±10.4	11.3±1.4	6.5±0.1
	6	113 ±9.9	11.8±1.6	6.6±0.1
Mytilus edulis (10 cm) Mytilus californianus	1	control	9.9±0.5	
	2	10 ±2.6	9.6±0.6	
Test length: 262 hrs	3	14 ±2.5	9.5±0.6	
Salinity: 31±1 ppt Dissolved oxygen: 8±1 ppm	4	17 ±3.5	9.6±0.6	
	5	24 ±3.2	9.6±0.8	
	6	44 ±5.3	9.7±0.9	
	7	68 ±8.6	9.6±0.9	
	8	99±15.0	9.6±0.8	
	9	113±11.9	9.6±0.9	

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Appendix A, Table AII, Experimental conditions (continued) 3

Species	Test Conc. No.	Suspended Kaolin gm/l	Temp. °C	pH
Molgula manhattensis Stylela montereyensis	1	control	12.3±0.2	7.6±0.1
	2	11 ±1.3	12.2±0.1	7.4±0.1
Test length: 288 hrs Salinity: 21.6±.9 ppt Dissolved oxygen: 8±1 ppm	3	15 ±1.8	12.2±0.2	7.2±0.1
	4	26 ±2.0	12.3±0.1	7.1±0.1
	5	38 ±3.0	12.3±0.1	6.8±0.1
	6	72 ±5.5	12.4±0.3	6.6±0.1
	7	102 ±8.5	12.4±0.1	6.3±0.1
Ascidia ceratodes	1	control	8.5±0.1	8.1±0.1
Test length: 136 hrs Salinity: 33±2 ppt Dissolved oxygen: 8.5±1 ppm	2	10 ±1.2	8.8±0.1	7.4±0.1
	3	12 ±2.4	8.5±0.2	7.4±0.1
	4	15 ±3.1	8.6±0.5	7.3±0.1
	5	18 ±2.9	8.6±0.5	7.3±0.1
	6	23 ±2.7	8.6±0.5	7.0±0.1
	7	25 ±1.8	8.6±0.1	7.0±0.1
	8	39 ±2.5	8.6±0.2	6.5±0.1
	9	68 ±2.9	8.5±0.3	6.5±0.1
	10	101 ±1.8	8.6±0.5	6.3±0.1
	11	109 ±5.6	8.6±0.4	6.3±0.1
Crangon nigromaculata	1	control	10.5±0.4	8.0±0.1
Test length: 397 hrs Salinity: 31±1 ppt Dissolved oxygen: 8.2±1 ppm	2	11 ±2.8	10.1±0.3	7.7±0.1
	3	15 ±2.5	10.2±0.3	7.7±0.1
	4	17 ±3.1	10.3±0.3	7.6±0.2
	5	25 ±5.5	10.5±0.3	7.4±0.2
	6	40 ±8.7	10.5±0.3	7.2±0.2

Appendix A, Table AII, Experimental conditions (continued) 4

Species	Test Conc. No.	Suspended Kaolin gm/l	Temp. °C	pH
Crangon nigromaculata (cont.)	7	63±12.3	10.1±0.2	6.9±0.2
	8	101±15.5	10.2±0.2	6.6±0.3
Crangon nigricauda	1	control	15.2±0.1	7.5±0.1
Test length: 246 hrs Salinity: 21±4 ppt Dissolved oxygen: 8±1 ppm	2	9 ±0.4	15.1±0.1	7.2±0.1
	3	17 ±0.6	15.1±0.1	7.0±0.1
	4	23 ±2.1	15.0±0.1	6.8±0.1
	5	35 ±1.4	15.1±0.1	6.6±0.1
	6	74 ±6.3	15.1±0.1	6.4±0.1
	7	117 ±7.7	15.2±0.1	6.0±0.1
Palaemon macrodactylus Anisogammarus confervicolus	1	control	11.5±1.4	7.7±0.2
	2	9 ±0.9	11.1±1.4	7.5±0.2
Test length: 269 hrs Salinity: 13.2±.5 ppt Dissolved oxygen: 8±1 ppm	3	12 ±1.5	11.1±1.4	7.4±0.2
	4	31 ±2.7	11.2±1.3	7.1±0.1
	5	102±10.4	11.3±1.4	6.5±0.1
	6	113 ±9.9	11.8±1.6	6.6±0.1
Cancer magister	1	control	10.5±0.5	8.1±0.1
Test length: 240 hrs Salinity: 32.4±1.5 ppt Dissolved oxygen: 8±1 ppm	2	10 ±3.5	10.3±0.3	7.8±0.2
	3	14 ±4.4	10.2±0.2	7.6±0.1
	4	26 ±8.0	10.2±0.2	7.4±0.1
	5	38±11.5	10.4±0.4	7.2±0.1

Appendix A, Table AII, Experimental conditions (continued) 5

Species	Test Conc. No.	Suspended Kaolin gm/l	Temp °C	pH
Neanthes succinea	1	control	11.5±1.4	7.7±0.2
Test length: 260 hrs Salinity: 13.2±.5 ppt Dissolved oxygen: 8.5±1 ppm	2	9 ±0.9	11.1±1.4	7.5±0.2
	3	31 ±2.7	11.2±1.3	7.1±0.1
	4	102±10.4	11.3±1.4	6.5±0.1
	5	113 ±9.9	11.8±1.6	6.6±0.1
Parophrys vetulus	1	control	15.2±0.1	7.5±0.1
Test length: 238 hrs Salinity: 21±4 ppt Dissolved oxygen: 8.5±1 ppm	2	9 ±0.4	15.1±0.1	7.2±0.1
	3	17 ±0.6	15.1±0.1	7.0±0.1
	4	23 ±2.1	15.0±0.1	6.8±0.1
	5	35 ±1.4	15.1±0.1	6.6±0.1
	6	74 ±6.3	15.1±0.1	6.4±0.1
	7	117 ±7.7	15.2±0.1	6.0±0.1
Cymatogaster aggregata (Bodega Bay specimens)	1	control	8.7±0.7	Not recorded
	2	12 ±3.6	8.6±0.5	
Test length: 26 hrs Salinity: 31±1 ppt Dissolved oxygen: 8±1 ppm	3	17 ±2.8	8.7±0.8	
	4	20 ±4.5	8.6±0.7	
	5	34 ±3.6	8.9±0.9	
	6	60 ±3.5	8.7±0.8	
	7	100 ±4.5	8.9±0.8	
Cymatogaster aggregata (S.F. Bay specimens)	1	control	12.9±3.3	7.5±0.2
	2	2 ±0.39	12.9±3.3	7.5±0.2
Test length: 201 hrs Salinity: 15.5±.8 ppt Dissolved oxygen: 8.4±1.p ppm	3	4 ±0.77	12.6±3.5	7.5±0.2

APPENDIX B

Animal Collection and Holding Data and Experimental Conditions for the Suspended Bentonite-Temperature Mortality Experiments

APPENDIX B. BENTONITE-TEMPERATURE MORTALITY EXPERIMENTS COLLECTION AND HOLDING DATA

TABLE BI. List of species tested; date, method and site of collection; and the salinity and temperature from which the animals were collected. Also presented is the length of time each species was maintained in the laboratory before testing began; the salinity at which they were held; the initial temperature and the final temperatures to which each was acclimated for testing. Temperature was changed from the initial to the final at a rate of 2°C every 2 days.

	Collection Data					Holding Data				
Species	date (1974)	method	site	sal. ppt	temp. °C	days in lab	sal. ppt	init. temp. °C	final warm temp. °C	final cool temp. °C
Parophrys vetulus	early July	otter trawl	San Bruno Shoal area	27-28	17-18	4-12	21-29	18	18	10
Mytilus edulis	early July	hand	Central S.F. Bay	27-30	18-20	2-12	21-29	18	18	10
Crangon nigricauda	early July	otter trawl	San Bruno Shoal area	27-28	17-18	4-12	21-29	18	18	10
Cymatogaster aggregata	early July	otter trawl	San Bruno Shoal area	27-28	17-18	4-12	21-29	18	18	
Cymatogaster aggregata	mid-July	otter trawl	San Bruno Shoal area	27-28	17-18	6-14	21-27	18		10
Neanthes succinea	mid-July	hand	Central S.F. Bay	27-30	18-20	6-14	21-29	18	18	10
Anisogammarus confervicolus	late July	dipnet	Mare Island area	16-22	18-22	12	21-29	18	18	10

APPENDIX B. BENTONITE-TEMPERATURE MORTALITY EXPERIMENTS EXPERIMENTAL CONDITIONS

TABLE BII. Summary of species tested, duration of the experiments, the salinity and dissolved oxygen common to all aquaria, the number of test aquaria used, and the suspended solids concentration, temperature and pH in each aquarium. Values are means ±1 standard deviation. Species grouped together in the table were tested simultaneously in the same aquaria.

Species	Test Conc. No.	Suspended solids gm/l	Temp. °C	pH
Parophrys vetulus	1	control	18.0±0.4	7.3±0.1
Mytilus edulis	2	8 ±0.8	17.9±0.3	7.3±0.1
Crangon nigricauda	3	11 ±0.7	17.9±0.3	7.3±0.1
Test length: 236 hrs Salinity: 27.1±.7 ppt Dissolved oxygen: 8.0±1.0 ppm 18°C	4	16 ±1.2	17.9±0.2	7.2±0.1
	5	24 ±1.9	17.9±0.2	7.3±0.1
	6	36 ±2.0	17.9±0.2	7.2±0.1
	7	54 ±2.3	17.9±0.2	7.2±0.1
Parophrys vetulus	1	control	10.1±0.7	7.4±0.1
Mytilus edulis	2	7 ±2.8	10.0±0.8	7.4±0.1
Crangon nigricauda	3	8 ±0.7	10.0±1.1	7.4±0.1
Test length: 236 hrs Salinity: 28.4±.7 ppt Dissolved oxygen: 8.2±1.0 ppm 10°C	4	13 ±1.2	10.2±0.6	7.4±0.1
	5	21 ±1.9	10.4±0.3	7.3±0.1
	6	36 ±4.0	10.0±0.8	7.3±0.1
	7	55 ±4.3	10.2±0.6	7.3±0.1

Species	Test Conc. No.	Suspended solids gm/l	Temp. °C	pH
Cymatogaster aggregata	1	control	17.8±0.2	7.5±0.1
Test length: 195 hrs Salinity: 26.7±1.5 ppt Dissolved oxygen: 8±1 ppm 18°C	2	0.4 ±.06	17.8±0.1	7.5±0.1
	3	0.6 ±.07	17.8±0.1	7.5±0.1
	4	1.0 ±.13	17.9±0.1	7.5±0.1
	5	1.6 ±.30	17.9±0.1	7.5±0.1
	6	3.2 ±.20	17.8±0.1	7.5±0.1
	7	3.6 ±.29	17.9±0.1	7.5±0.1
Cymatogaster aggregata	1	control	10.9±1.2	7.5±0.1
Test length: 181 hrs Salinity: 25.2±5 ppt Dissolved oxygen: 8.3±1.0 ppm 10°C	2	0.4 ±.07	10.5±1.0	7.6±0.1
	3	0.7 ±.08	10.4±1.0	7.6±0.1
	4	1.1 ±.16	10.4±0.6	7.6±0.1
	5	1.7 ±.42	10.6±0.9	7.6±0.1
	6	3.3 ±.67	10.6±1.4	7.6±0.1
	7	4.1 ±.88	10.5±1.4	7.6±0.1
Neanthes succinea	1	control	18.4±0.3	7.5±0.1
Anisogammarus confervicolus	2	6 ±0.9	18.5±0.3	7.5±0.1
Test length: 237 hrs Salinity: 26.2±1.3 ppt Dissolved oxygen: 8.0±1.0 ppm 18°C	3	9 ±1.9	18.5±0.3	7.4±0.1
	4	14 ±1.4	18.5±0.3	7.4±0.1
	5	23 ±2.0	18.6±0.4	7.3±0.1
	6	35 ±2.8	18.6±0.3	7.3±0.1
	7	53 ±3.8	18.7±0.3	7.2±0.1

Species	Test Conc. No.	Suspended solids gm/l	Temp. °C	pH
Neanthes succinea	1	control	10.9±0.9	7.6±0.1
Anisogammarus confervicolus	2	7 ±1.1	10.5±0.1	7.6±0.1
Test length: 237 hrs Salinity: 24.2±5.9 ppt Dissolved oxygen: 8.2±1.0 ppm 10°C	3	10 ±1.0	10.3±0.5	7.6±0.1
	4	15 ±1.4	10.2±0.5	7.5±0.1
	5	22 ±2.1	10.2±0.5	7.5±0.1
	6	33 ±2.6	10.3±0.5	7.4±0.1
	7	54 ±3.6	10.6±1.1	7.3±0.1

APPENDIX C

Animal Collection and Holding Data and Experimental Conditions for the Suspended Bentonite-Dissolved Oxygen Mortality Experiments.

APPENDIX C. BENTONITE-DISSOLVED OXYGEN MORTALITY EXPERIMENTS COLLECTING AND HOLDING DATA

TABLE CI. List of species tested; date, method and site of collection; and the salinity and temperature from which the animals were collected. Also presented is the length of time each species was maintained in the laboratory before testing began, and the salinity and temperature at which each was held.

Species	COLLECTION DATA					HOLDING DATA		
	date (1974)	method	site	salinity ppt	temp. °C	days held in lab	salinity ppt	temp. °C
Mytilus edulis	mid-Aug.	hand	Central S.F. Bay	27-30	18-20	15	25-29	18
Morone saxatilis		HATCHERY FISH					25-29	18
Cymatogaster aggregata	Sept.	otter trawl	San Bruno Shoal area	27-30	18-20	8-22	25-29	18
Crangon nigricauda	early Sept.	otter trawl	San Bruno Shoal area	27-30	18-20	10-15	25-29	18
Synidotea laticauda	late Aug.	dipnet	Central S.F. Bay	16-22	18-20	8-16	25-29	18
Neanthes succinea	late Aug.	hand	Central S.F. Bay	27-30	18-20	8-22	25-29	18

APPENDIX C. BENTONITE-DISSOLVED OXYGEN MORTALITY EXPERIMENTS EXPERIMENTAL CONDITIONS

TABLE CII. Summary of species tested, duration of the experiments, the salinity common to all aquaria, the number of test aquaria used, and the suspended solids concentration, temperature, pH and dissolved oxygen in each aquarium. Values presented are means ±1 standard deviation. Species grouped together in the table were tested simultaneously in the same aquaria.

Species	Test Conc. No.	Suspended solids gm/l	D.O. ppm	Temp. °C	pH
Mytilus edulis	1	Control	7.8±0.3	18.0±0.1	7.6±0.1
Test length: 240 hrs Salinity: 24.6±.7 ppt 5 ppm D.O.	2	6±0.4	5.1±0.3	17.9±0.1	7.6±0.1
	3	8±0.9	6.1±1.0	17.9±0.1	7.5±0.1
	4	15±2.0	5.3±1.2	17.9±0.2	7.4±0.1
	5	21±1.4	4.9±0.4	18.0±0.2	7.4±0.1
	6	35±3.6	5.0±0.6	17.9±0.2	7.3±0.1
	7	48±3.5	4.8±0.4	18.0±0.1	7.3±0.1
Mytilus edulis	1	Control	7.7±0.4	18.1±0.2	7.6±0.1
Test length: 240 hrs Salinity: 26.6±4 ppt 2 ppm D.O.	2	6±0.7	2.2±0.2	18.0±0.2	7.6±0.1
	3	10±0.6	2.2±0.5	18.0±0.2	7.6±0.1
	4	14±1.3	2.1±0.2	18.1±0.3	7.5±0.1
	5	21±1.7	2.0±0.2	18.1±0.3	7.4±0.1
	6	31±3.2	2.2±0.3	18.1±0.3	7.4±0.1
	7	46±5.4	2.1±0.3	18.1±0.2	7.3±0.1

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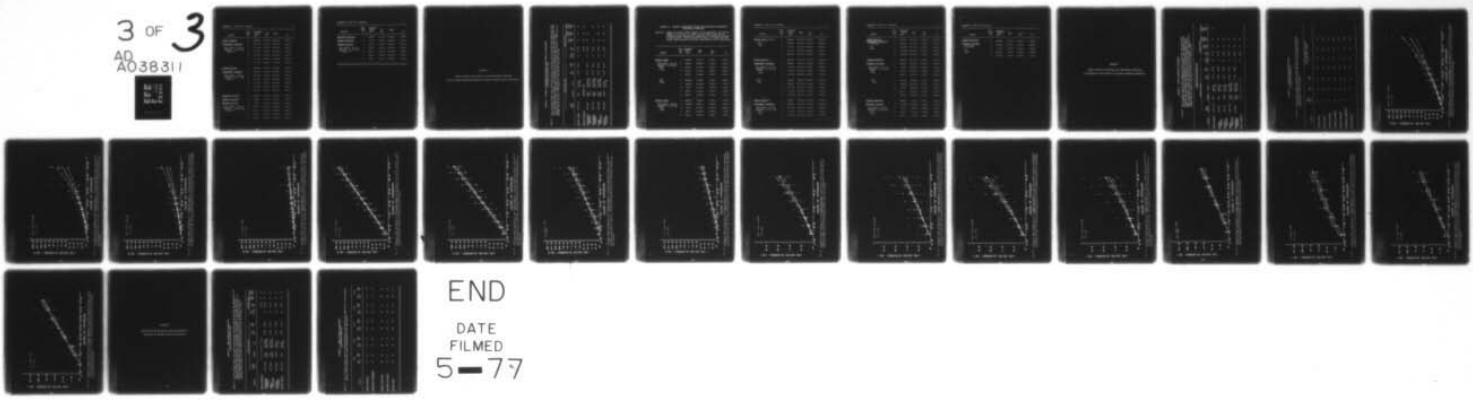

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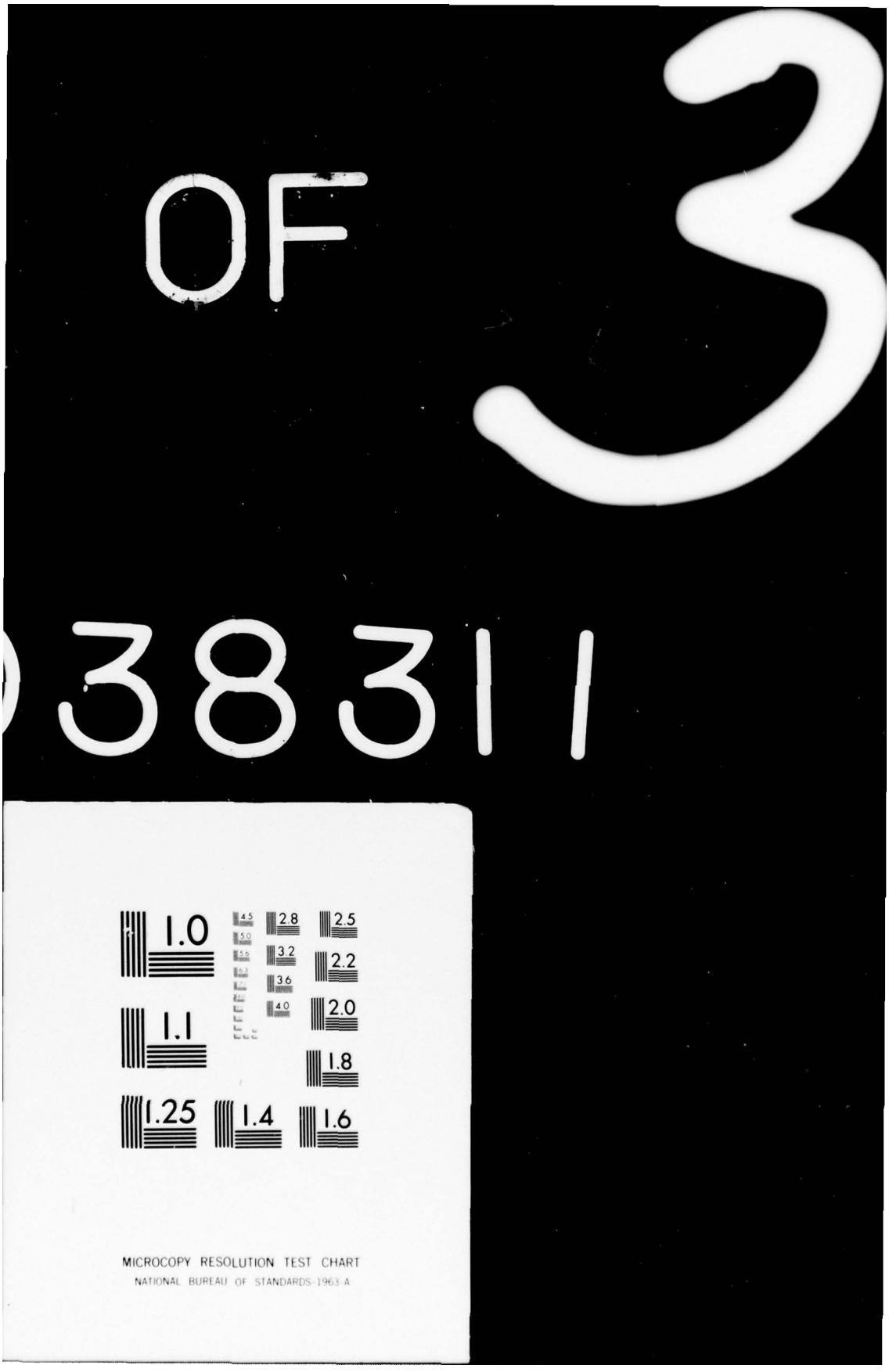
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Species	Test Conc. No.	Suspended solids gm/l	D.O. ppm	Temp. °C	pH
Morone saxatilis	1	control	7.4±0.3	18.2±0.3	7.5±0.1
Cymatogaster aggregata	2	0.2±.05	5.5±1.5	18.1±0.3	7.5±0.1
Test length: 240 hrs	3	0.3±.05	5.0±0.2	18.0±0.3	7.5±0.1
Salinity: 24.6±.5 ppt 5 ppm D.O.	4	0.5±.06	5.2±0.3	18.1±0.3	7.5±0.1
	5	0.6±.07	5.2±0.2	18.2±0.2	7.5±0.1
	6	1.1±.14	5.0±0.2	17.9±0.2	7.5±0.1
	7	2.0±.28	5.0±0.2	17.9±0.2	7.5±0.1
Morone saxatilis	1	control	7.6±0.4	18.3±0.2	7.5±0.1
Cymatogaster aggregata	2	0.6±.06	2.1±0.5	18.2±0.1	7.6±0.1
Test length: 240 hrs	3	0.9±.09	2.3±0.4	18.1±0.1	7.6±0.1
Salinity: 24.5±.9 ppt 2 ppm D.O.	4	1.3±.11	2.3±0.4	18.1±0.1	7.6±0.1
	5	2.5±.19	3.0±2.1	18.1±0.1	7.6±0.1
	6	4.2±.51	2.4±0.8	18.1±0.1	7.6±0.1
	7	6.5±.35	2.8±1.5	18.2±0.2	7.6±0.1
Synidotea laticauda	1	control	7.8±0.4	17.9±0.1	7.6±0.1
Crangon nigricauda	2	7±0.5	5.1±1.0	17.8±0.1	7.6±0.1
Neanthes succinea	3	9±0.6	5.8±2.2	17.8±0.1	7.6±0.1
Test length: 240 hrs	4	15±1.1	5.3±0.4	17.8±0.1	7.5±0.1
Salinity: 24.5±1.0 ppt 5 ppm D.O.	5	25±3.1	5.3±0.4	17.8±0.1	7.5±0.1
	6	35±4.1	5.1±0.4	17.8±0.1	7.4±0.1
	7	47±4.6	5.1±0.2	17.9±0.1	7.4±0.1

Species	Test Conc. No.	Suspended solids gm/l	D.O. ppm	Temp. °C	pH
Synidotea laticauda	1	Control	7.6±0.4	18.3±0.1	7.6±0.1
Crangon nigricauda	2	8±1.0	2.2±0.4	18.2±0.1	7.6±0.1
Neanthes succinea	3	11±0.9	2.3±0.4	18.2±0.1	7.6±0.1
Test length: 240 hrs Salinity: 24.3±.7 ppt 2 ppm D.O.	4	15±1.2	2.6±1.2	18.2±0.2	7.6±0.1
	5	21±2.8	2.3±0.4	18.1±0.2	7.5±0.1
	6	33±2.6	2.8±1.2	18.1±0.1	7.5±0.1
	7	49±3.3	2.2±0.5	18.1±0.3	7.4±0.1

APPENDIX D

Animal Collection and Holding Data and Experimental Conditions for the Suspended Bentonite-Temperature-Dissolved Oxygen Mortality Experiments.

APPENDIX D. BENTONITE-TEMPERATURE-DISSOLVED OXYGEN MORTALITY EXPERIMENTS COLLECTION AND HOLDING DATA

TABLE DI. List of species tested, date, method and site of collection, and the salinity and temperature from which the animals were collected. Also presented is the length of time each species was maintained in the laboratory before testing began; the salinity at which they were held; the initial temperature and the final temperature to which each was acclimated before testing. Temperature was changed from the initial to the final at a rate of 2°C every two (2) days.

	COLLECTION DATA					HOLDING DATA				
	date (1974)	method	site	sal. ppt	temp. °C	days in lab	sal. ppt	init. temp. °C	final warm temp. °C	final cool temp. °C
Mytilus edulis	early Oct.	hand	Central S.F. Bay	27-30	18-20	16	24-28	18	18	10
Morone saxatilis		HATCHERY FISH					24-28	18	18	10
Cymatogaster aggregata	mid-Sept.	otter trawl	San Bruno Shoal area	27-30	18-20	10	24-28	18	18	
Cymatogaster aggregata	early Oct.	otter trawl	San Bruno Shoal area	27-30	18-20	13	24-28	18		10
Crangon nigricauda	late Sept.	otter trawl	San Bruno Shoal area	27-30	18-20	10	24-28	18	18	10
Synidotea laticauda	late Sept.	dipnet	Central S.F. Bay	25-27	18-20	11	24-28	18	18	10

APPENDIX D. BENTONITE-TEMPERATURE-DISSOLVED OXYGEN MORTALITY EXPERIMENTS EXPERIMENTAL CONDITIONS

TABLE DII. Summary of species tested, duration of the experiments, the salinity common to all aquaria, the number of test aquaria used, and the suspended solids concentration, temperature, pH and dissolved oxygen in each aquarium. Values presented are means ±1 standard deviation. Species listed together were tested simultaneously in the same aquaria.

Species	Test Conc. No.	Suspended solids gm/l	Temp. °C	D.O. ppm	pH
Mytilus edulis	1	control	17.7±0.1	7.4±0.6	7,8±0.1
Test length: 240 hrs	2	control	16.9±2.5	5.1±0.5	7.7±0.1
Salinity: 25.1±4 ppt					
18°C	3	11±2.8	17.7±0.1	6.1±1.2	7.6±0.1
5 ppm					
	4	23±2.6	17.7±0.2	4.9±0.3	7.6±0.1
	5	51±6.9	17.8±0.1	4.7±0.7	7.4±0.1
18°C	1	control	17.7±0.1	7.4±0.6	7.8±0.1
2 ppm					
	2	control	16.8±2.6	2.2±0.4	7.7±0.1
	3	7±1.6	17.7±0.2	2.1±0.3	7.7±0.1
	4	20±1.4	17.7±0.1	3.0±1.8	7.6±0.1
	5	54±5.5	17.8±0.2	2.1±0.2	7.5±0.1
Mytilus edulis	1	control	10.6±0.1	8.7±0.6	7.8±0.1
Test length: 240 hrs	2	control	102±0.2	5.4±0.3	7.7±0.1
Salinity: 25.7±2 ppt					
10°C	3	7±1.0	10.3±0.1	5.2±0.6	7.8±0.1
5 ppm					
	4	15±1.2	10.3±0.1	5.2±0.3	7.7±0.1
	5	52±7.8	10.4±0.1	4.9±0.4	7.5±0.1

Species	Test Conc. No.	Suspended solids gm/l	Temp. °C	D.O. ppm	pH
Mytilus edulis (contd.)	1	control	10.6±0.1	8.7±0.6	7.8±0.1
10°C 2 ppm	2	control	10.4±0.1	2.3±0.3	7.8±0.1
	3	8 ±0.9	10.3±0.1	2.3±0.2	7.8±0.1
	4	19 ±3.5	10.3±0.1	2.1±0.2	7.7±0.1
	5	48±10.9	10.4±0.1	2.0±0.2	7.6±0.1
Morone saxatilis	1	control	18.5±0.2	6.9±1.3	7.7±0.1
Cymatogaster aggregata	2	control	16.9±2.6	5.2±0.6	7.6±0.1
Test length: 240 hrs	3	1.1±0.11	18.4±0.2	5.1±0.3	7.7±0.1
Salinity: 25.1±.5 ppt 18°C 5 ppm	4	2.4±0.24	18.4±0.2	5.0±0.3	7.7±0.1
	5	6.5±0.61	18.4±0.2	5.2±0.2	7.7±0.1
18°C 2 ppm	1	control	18.5±0.2	6.9±1.3	7.7±0.1
	2	control	17.0±2.4	2.0±0.2	7.6±0.1
	3	0.7±0.15	18.4±0.2	2.1±0.2	7.8±0.1
	4	1.9±0.41	18.4±0.2	2.2±0.2	7.8±0.1
	5	7.1±0.50	18.5±0.1	2.1±0.2	7.9±0.1
Morone saxatilis	1	control	10.9±0.1	8.4±0.6	7.7±0.1
Cymatogaster aggregata	2	control	10.2±0.1	5.3±0.4	7.6±0.1
Test length: 240 hrs	3	0.9±0.07	10.9±0.1	5.4±0.4	7.8±0.1
Salinity: 25.8±.3 ppt 10°C 5 ppm	4	2.9±0.21	10.3±0.1	4.8±0.5	7.7±0.1
	5	5.4±0.56	10.1±0.1	5.2±0.1	7.7±0.1

Species	Test Conc. No.	Suspended Solids gm/l	Temp. °C	D.O. ppm	pH
Morone saxatilis	1	control	10.9±0.1	8.4±0.6	7.7±0.1
Cymatogaster aggregata (continued)	2	control	10.3±0.1	2.3±0.2	7.7±0.1
10°C 2 ppm	3	0.6±0.06	10.5±0.1	2.5±0.4	7.8±0.1
	4	1.6±0.05	10.2±0.1	2.2±0.2	7.8±0.1
	5	6.6±0.36	10.6±0.1	2.8±0.3	7.8±0.1
Crangon nigricauda	1	control	18.5±0.2	8.2±0.3	7.8±0.1
Synidotea laticauda	2	control	16.9±2.5	5.1±0.5	7.7±0.1
Test length: 240 hrs Salinity: 25.1±.5 ppt 18°C 5 ppm	3	9±0.8	18.5±0.2	5.2±0.2	7.8±0.1
	4	17±1.2	18.5±0.2	5.2±0.2	7.7±0.1
	5	44±6.2	18.5±0.2	5.1±0.3	7.6±0.1
18°C 2 ppm	1	control	18.5±0.2	8.2±0.3	7.8±0.1
	2	control	16.8±2.6	2.2±0.4	7.7±0.1
	3	9±0.4	18.5±0.2	2.3±0.2	7.8±0.1
	4	22±1.7	18.5±0.2	2.3±0.2	7.7±0.1
	5	47±7.5	18.5±0.2	2.2±0.1	7.6±0.1
Crangon nigricauda	1	control	9.7±0.2	8.4±2.4	7.9±0.1
Synidotea laticauda	2	control	10.2±0.2	5.4±0.3	7.7±0.1
Test length: 240 hrs Salinity: 25.5±.8 ppt 10°C 5 ppm	3	9±1.1	9.3±0.2	5.3±0.8	7.9±0.1
	4	18±1.4	9.1±0.2	5.2±0.5	7.8±0.1
	5	45±6.1	9.5±0.2	5.4±0.6	7.7±0.1

Species	Test Conc. No.	Suspended Solids gm/l	Temp. °C	D.O. ppm	pH
Crangon nigricauda	1	control	9.7±0.2	8.4±2.4	7.9±0.1
Synidotea laticauda (continued)	2	control	10.4±0.1	2.3±0.3	7.8±0.1
	3	7 ±1.2	9.2±0.2	2.1±0.2	7.9±0.1
10°C 2 ppm	4	23±12.0	9.1±0.2	2.5±1.0	7.8±0.1
	5	46 ±6.6	9.4±0.2	2.1±0.2	7.7±0.1

APPENDIX E

Animal Collection and Holding Data, Experimental Conditions and Responses of Each Species in the Oxygen Consumption Experiments

APPENDIX E. OXYGEN CONSUMPTION EXPERIMENTS

Table EI. List of species tested; date, method and site of collection, and the salinity and temperature from which the animals were collected. Also presented is the length of time each species was maintained in the laboratory before testing began; the salinity at which they were held; the initial temperature and the final temperature to which each was acclimated before testing. Temperature was changed from the initial to the final at a rate of 2°C every two days.

COLLECTION AND HOLDING DATA

	Collection Data					Holding Data				
Species	date (1974)	Method	site	sal. ppt	temp. °C	days in lab	sal. ppt	init. temp. °C	final warm temp. °C	final cool temp. °C
Mytilus edulis	early July	hand	Central S.F. Bay	27-30	18-20	10	21-29	18	18	11
Neanthes succinea	mid-July	hand	Central S.F. Bay	27-30	18-20	8	21-29	18	18	
Synidotea laticauda	late Aug.	dipnet	Central S.F. Bay	27-30	18-20	10	25-29	18	18	
Crangon nigricauda	early Sept.	otter trawl	San Bruno Shoal area	27-30	18-20	14	25-29	18	18	
Cymatogaster aggregata	mid-Sept.	otter trawl	San Bruno Shoal area	27-30	18-20	12	25-29	18	18	
Morone saxatilis		H A T C H E R Y F I S H				60	25-29	18	18	

APPENDIX E. OXYGEN CONSUMPTION EXPERIMENTS
EXPERIMENTAL CONDITIONS

Table EII. List of species tested, duration of the experiments, salinity and temperature at which the experiments were conducted, and the initial suspended bentonite concentration in each test vessel.

Species	Test length hrs.	sal. ppt.	temp. °C	Initial Suspended Bentonite Concentrations - gm/l 1	2	3	4
Mytilus edulis	12	29	11	0.0	5.5	13.0	30.0
Mytilus edulis	12	29	18	0.0	5.5	13.0	30.0
Neanthes succinea	12	28	18	0.0	5.5	13.0	30.0
Synidotea laticauda	12	28	18	0.0	5.5	13.0	30.0
Crangon nigricauda	8	28	18	0.0	1.0	3.0	10.0
Cymatogaster aggregata	4	24	18	0.0	1.0	3.0	10.0
Morone saxatilis	4	24	18	0.0	2.0	4.0	10.0

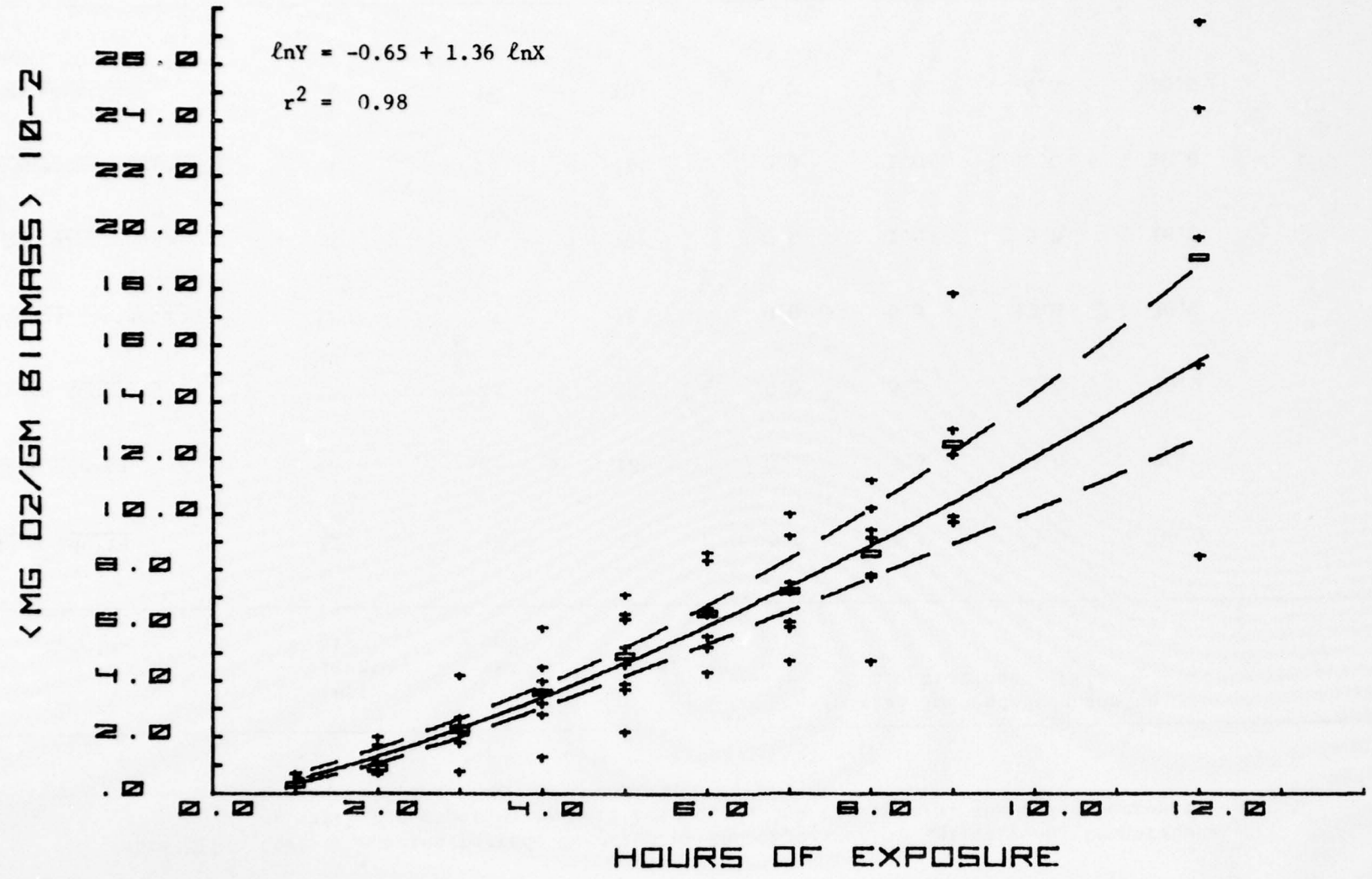

Fig. E1. Cumulative oxygen consumption of 19mm Mytilus edulis in clear water at 11°C and 29‰ salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval around that estimate.

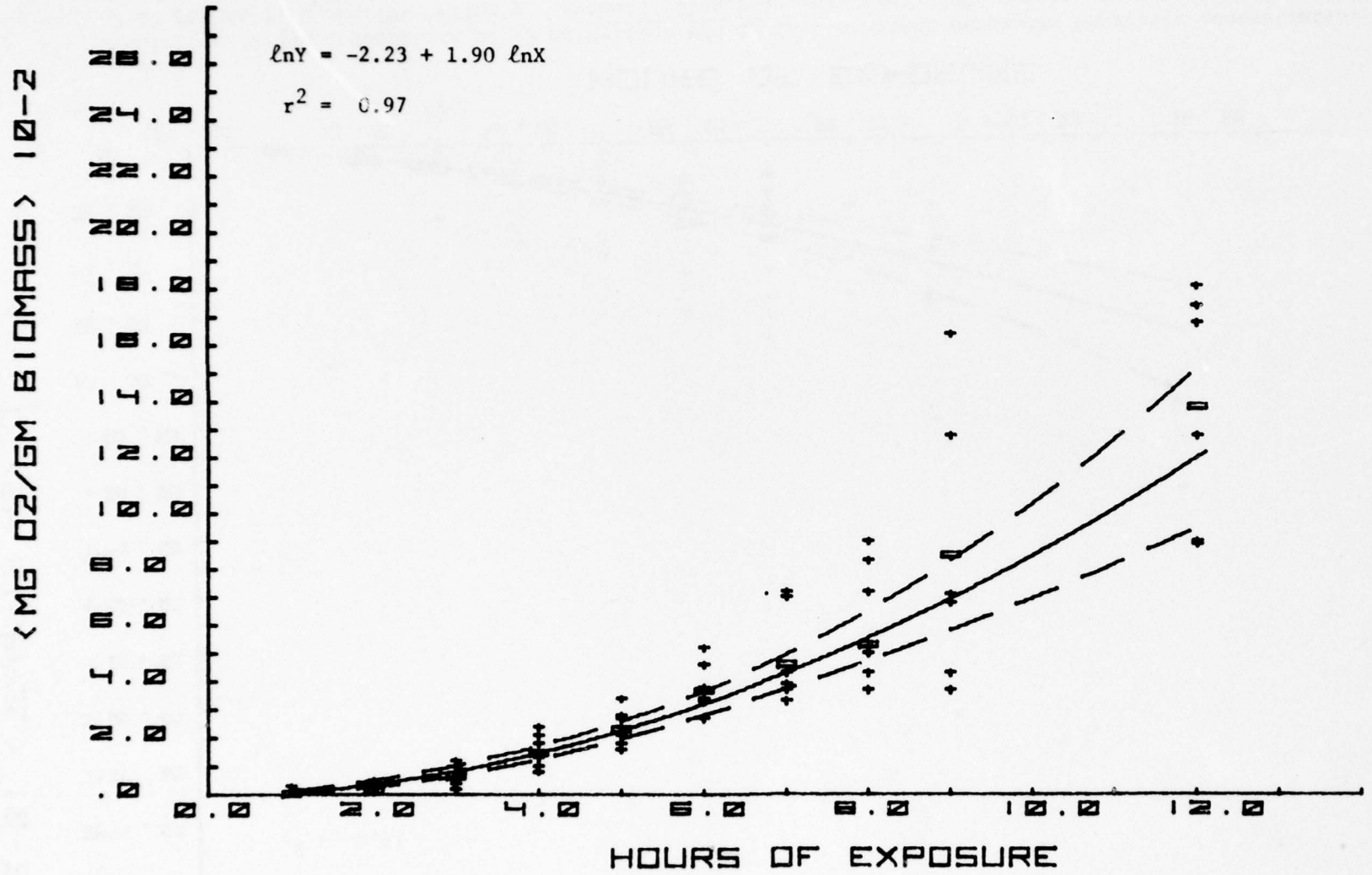

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Fig. E2. Cumulative oxygen consumption of 19mm Mytilus edulis in a starting suspended bentonite concentration of 5.5 gm/l at 11°C and 29% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval around that estimate.

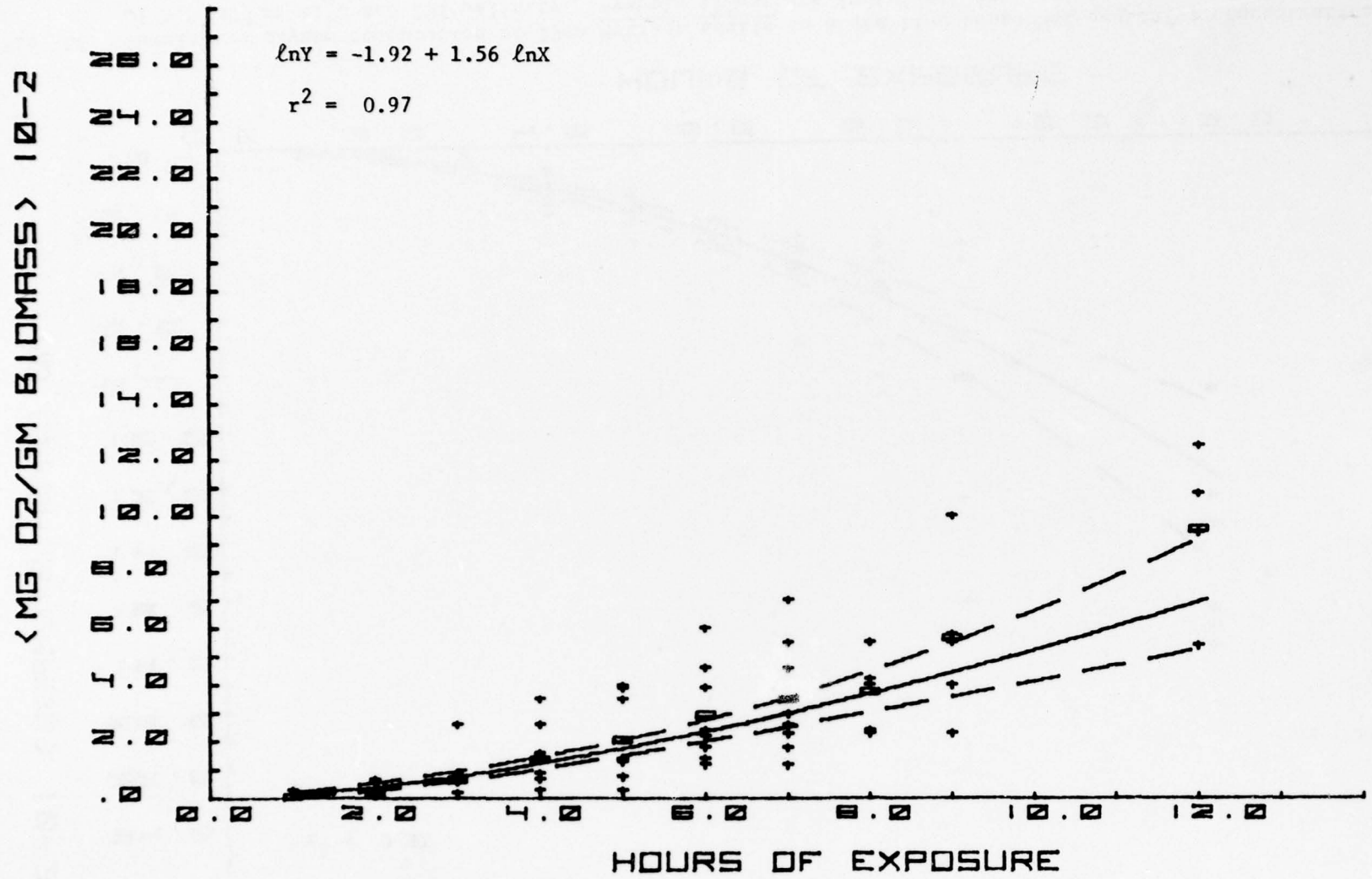

Fig. E3. Cumulative oxygen consumption of 19 mm Mytilus edulis in a starting suspended bentonite concentration of 13 gm/l at 11°C and 29% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

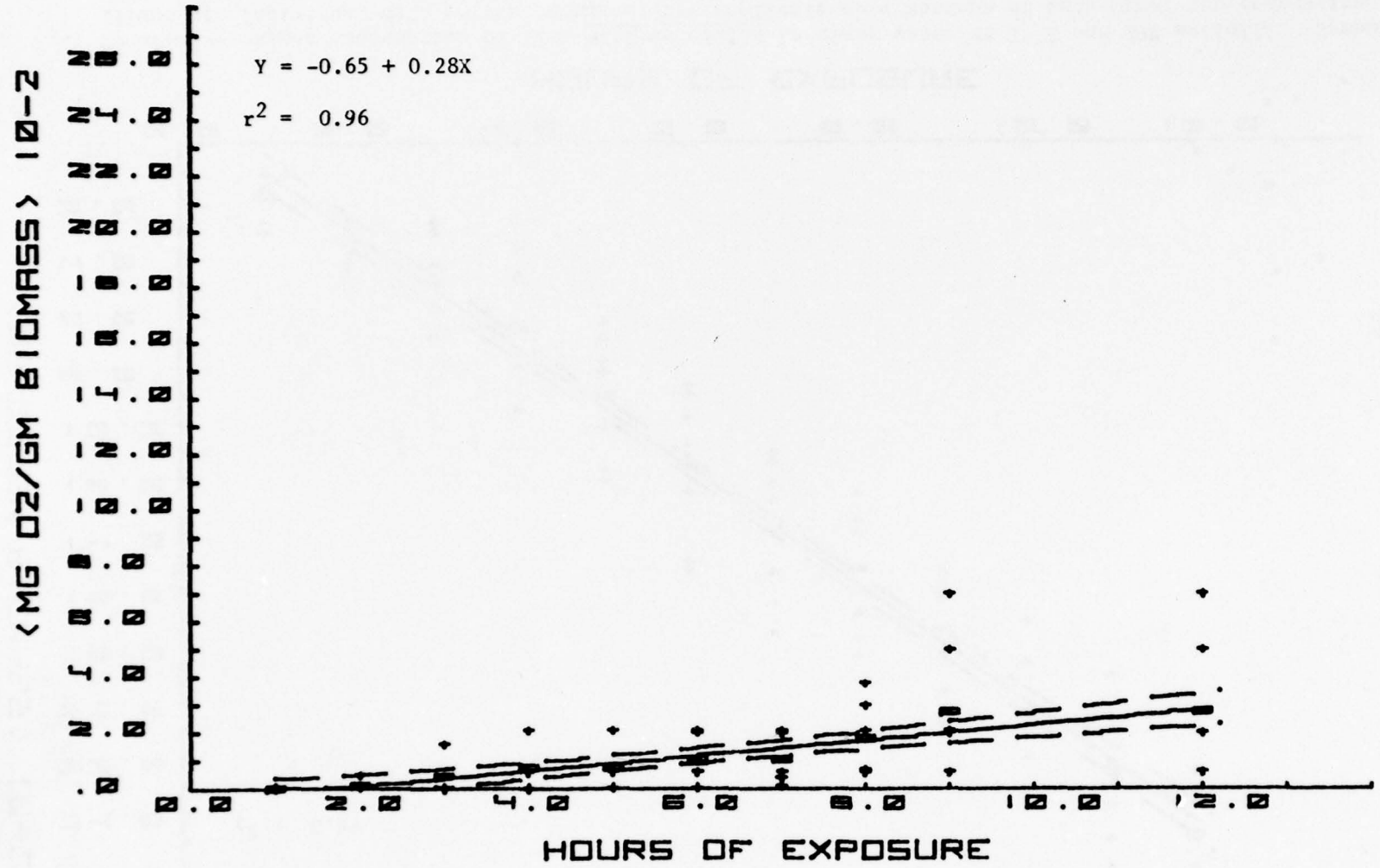

Fig. E4. Cumulative oxygen consumption of 19mm Mytilus edulis in a starting suspended bentonite concentration of 30 gm/l at 11°C and 29% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

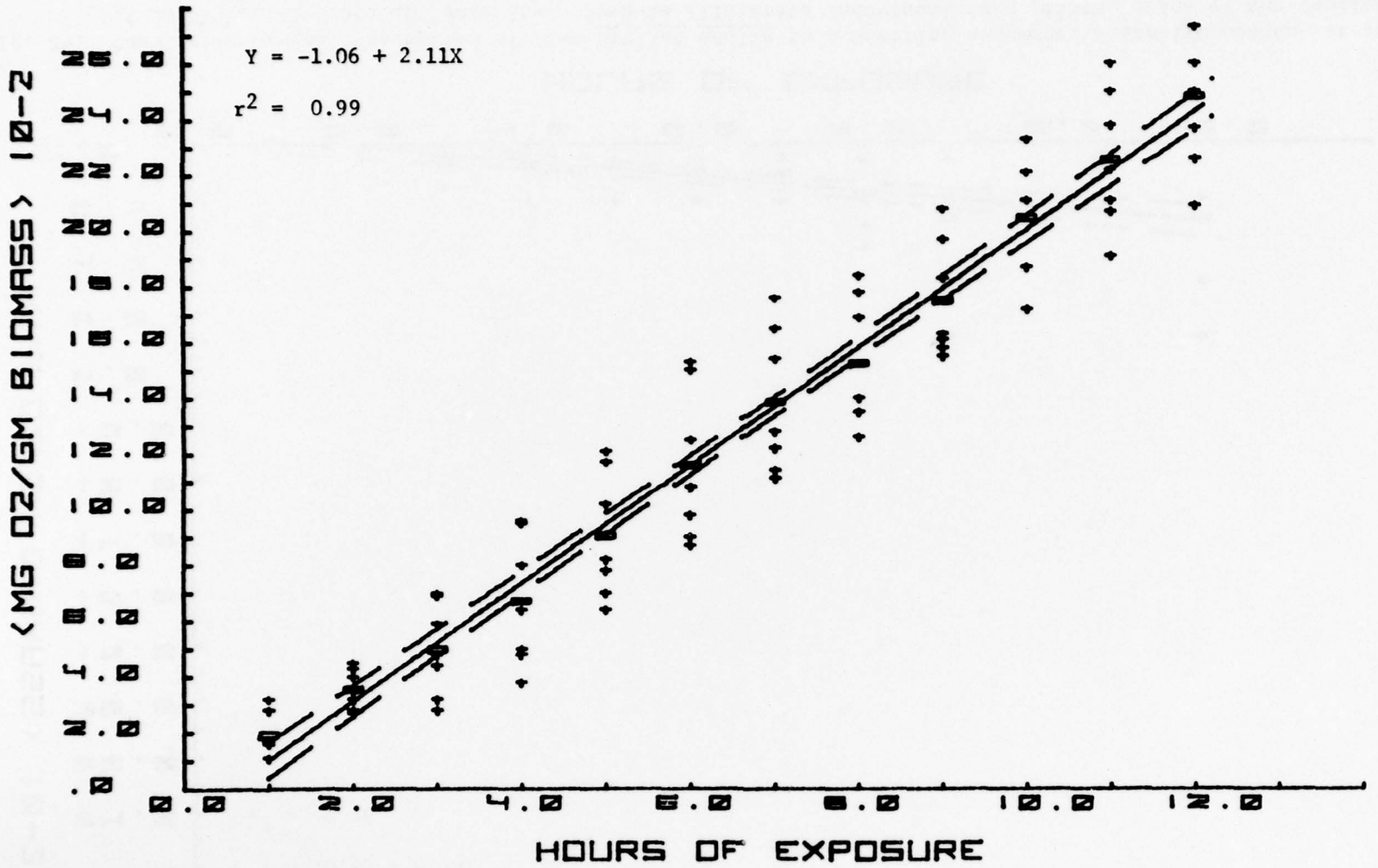

Fig. E5. Cumulative oxygen consumption of 22mm Mytilus edulis in clear water at 18°C and 29% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

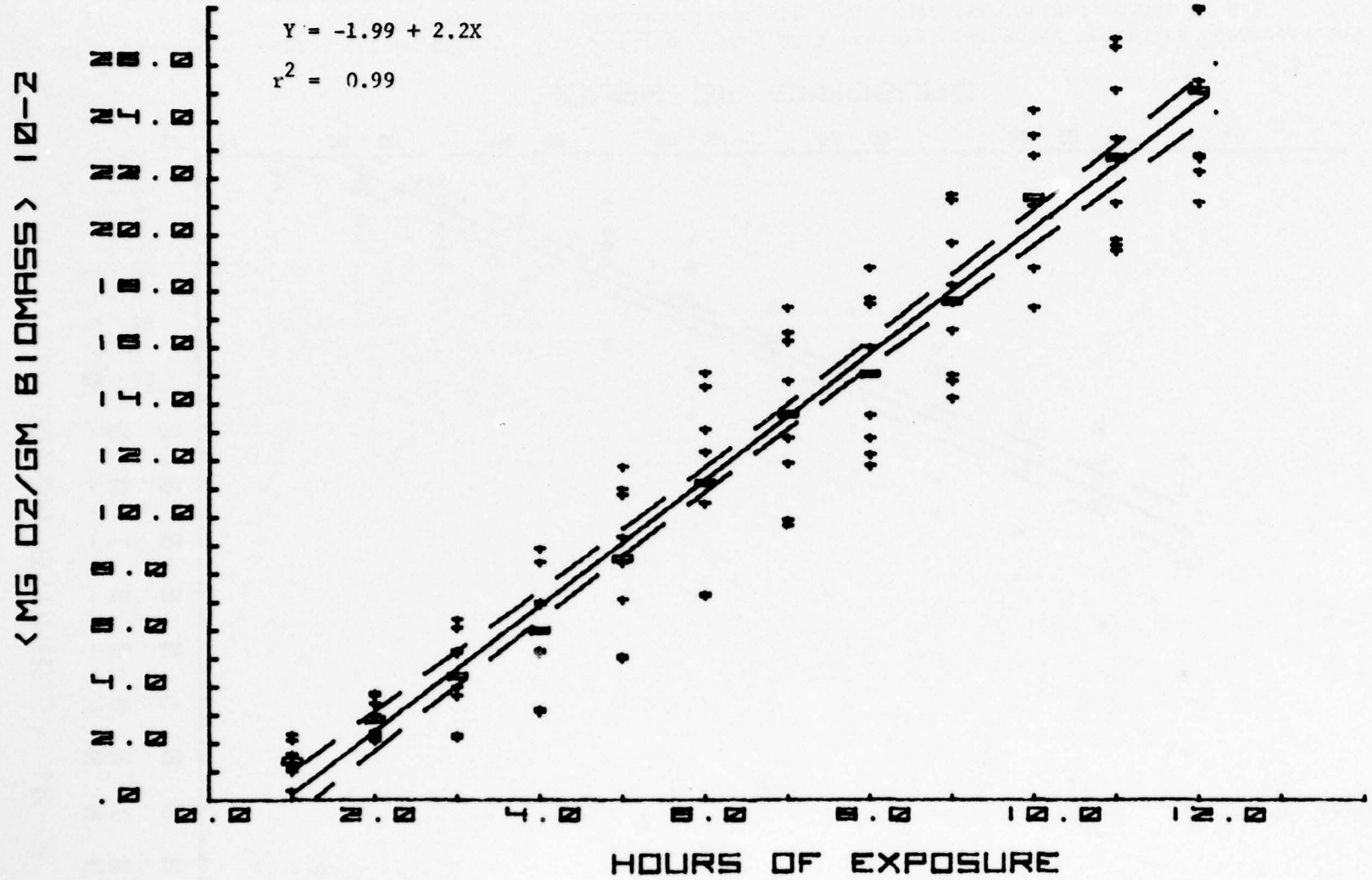

Fig. E6. Cumulative oxygen consumption of 22mm Mytilus edulis in a starting suspended bentonite concentration of 5.5 gm/l at 18°C and 29% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

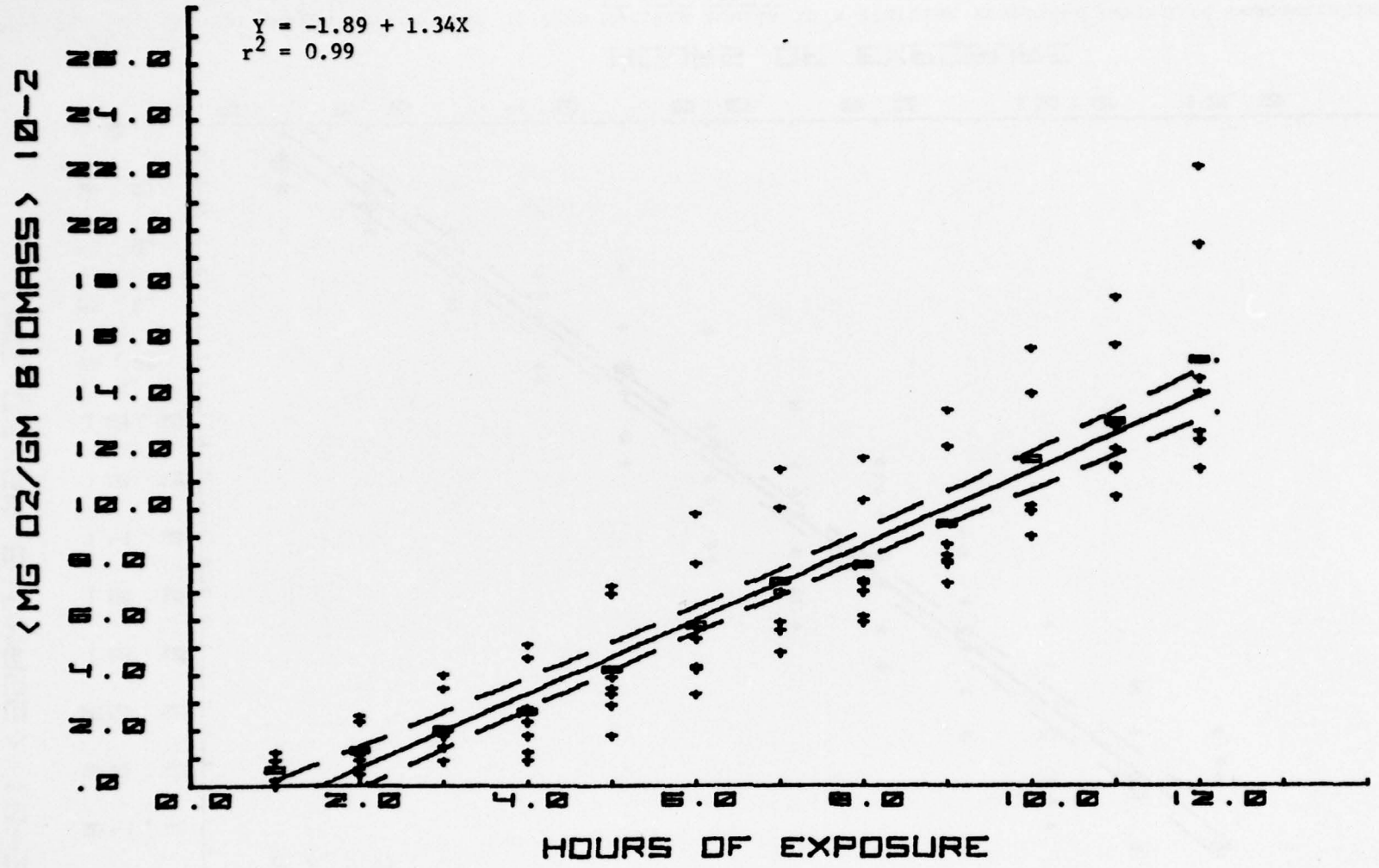

Fig. E7. Cumulative oxygen consumption of 22mm Mytilus edulis in a starting suspended bentonite concentration of 13 gm/l at 18°C and 29% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

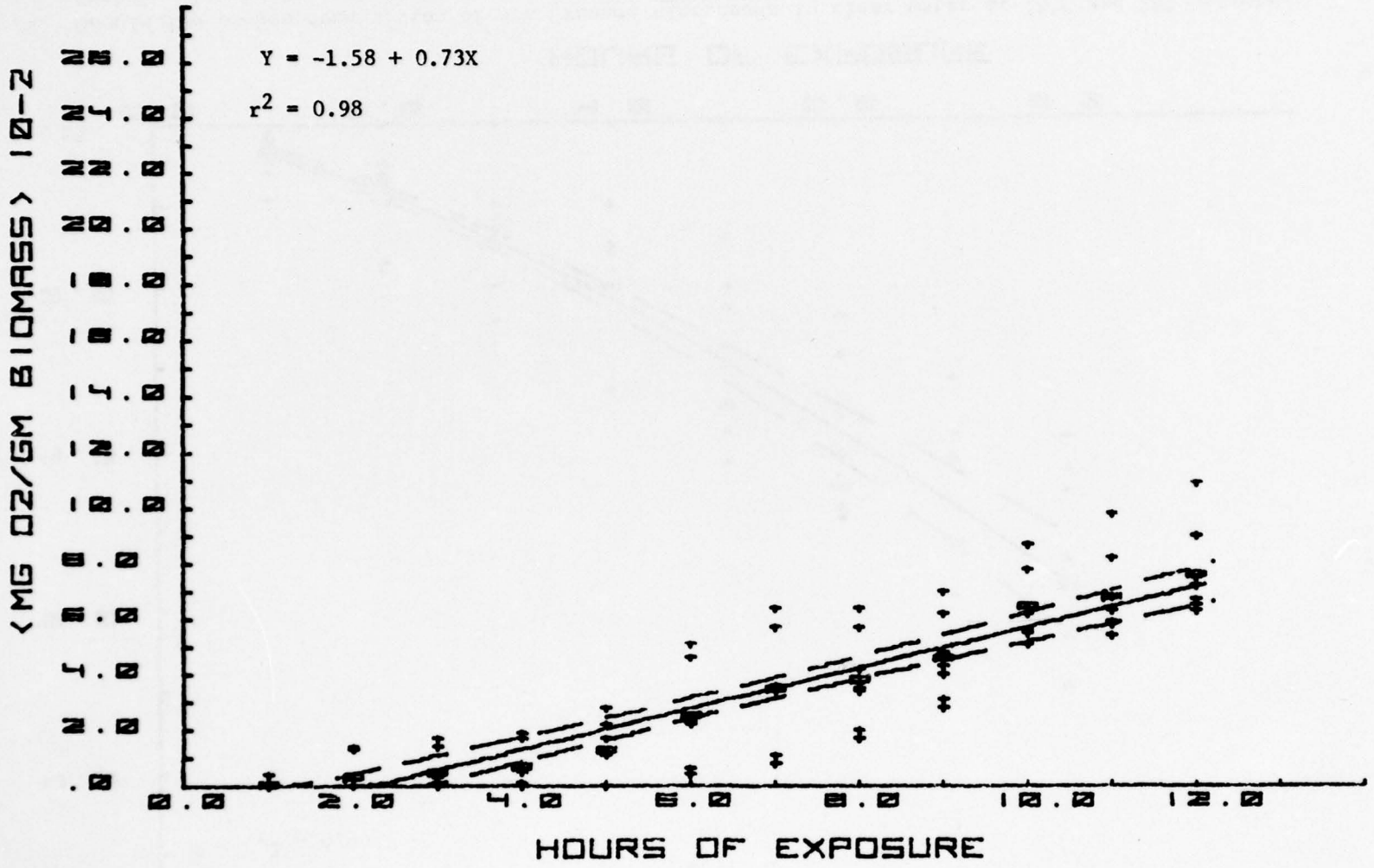

Fig. E8. Cumulative oxygen consumption of 22mm Mytilus edulis in a starting suspended bentonite concentration of 30 gm/l at 18°C and 29% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

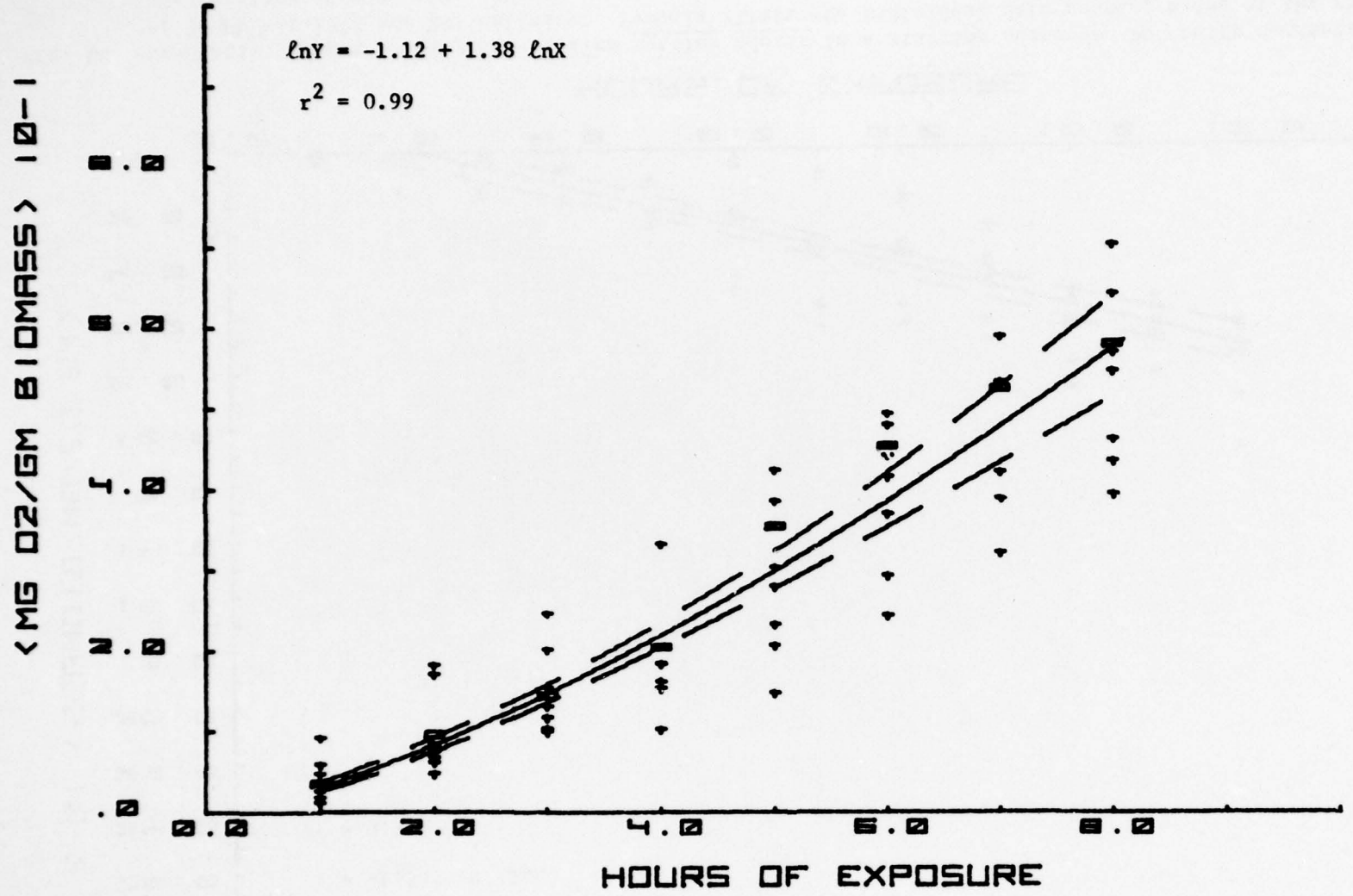

Fig. E9. Cumulative oxygen consumption of 4cm Crangon nigricauda in clear water at 18°C and 28% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

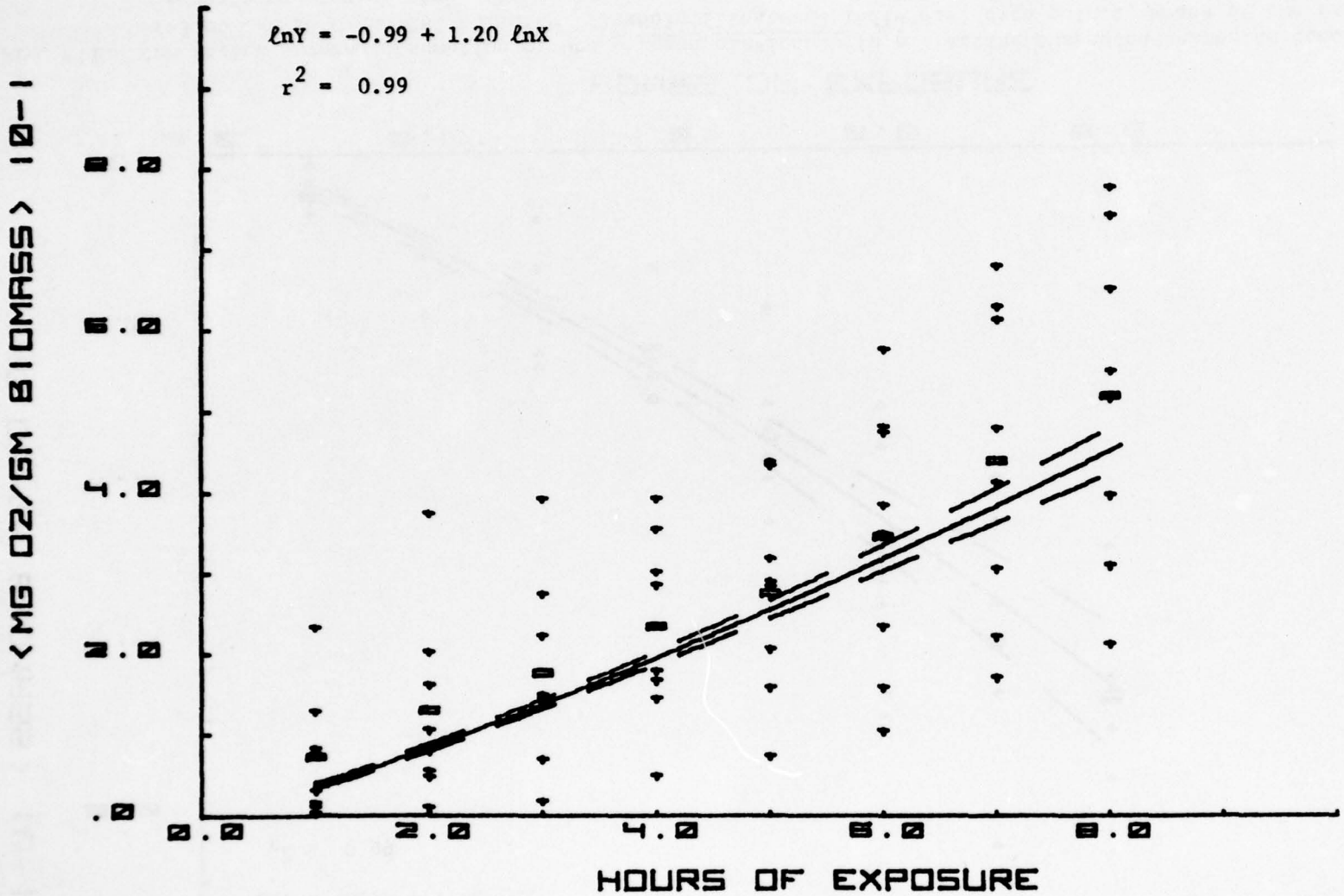

Fig. E10. Cumulative oxygen consumption of 4cm Crangon nigricauda in a starting suspended bentonite concentration of 1 gm/l at 18°C and 28% salinity. Symbols illustrate individual data points means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

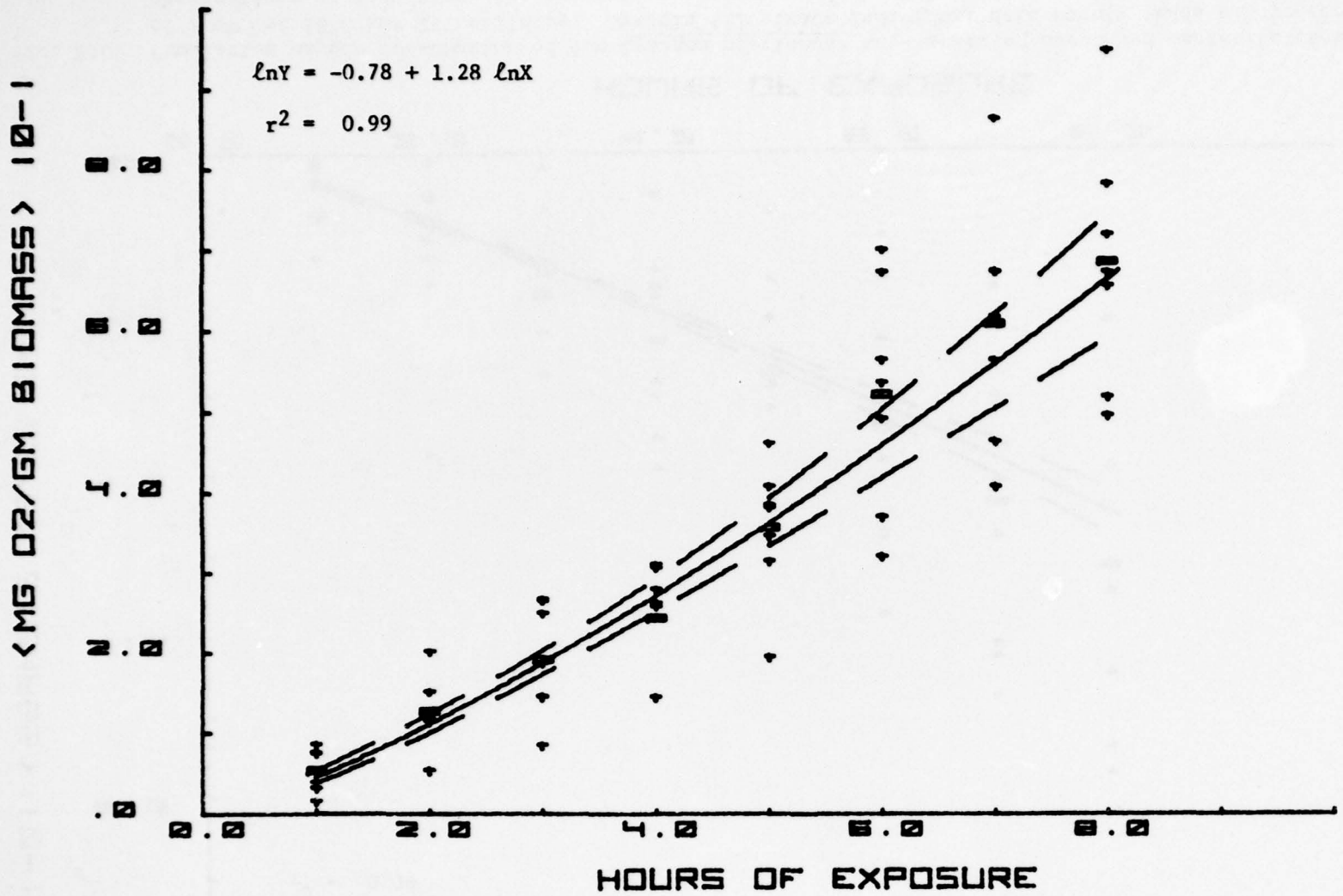

Fig. E11. Cumulative oxygen consumption of 4cm Crangon nigricauda in a starting suspended bentonite concentration of 3 gm/l at 18°C and 28% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

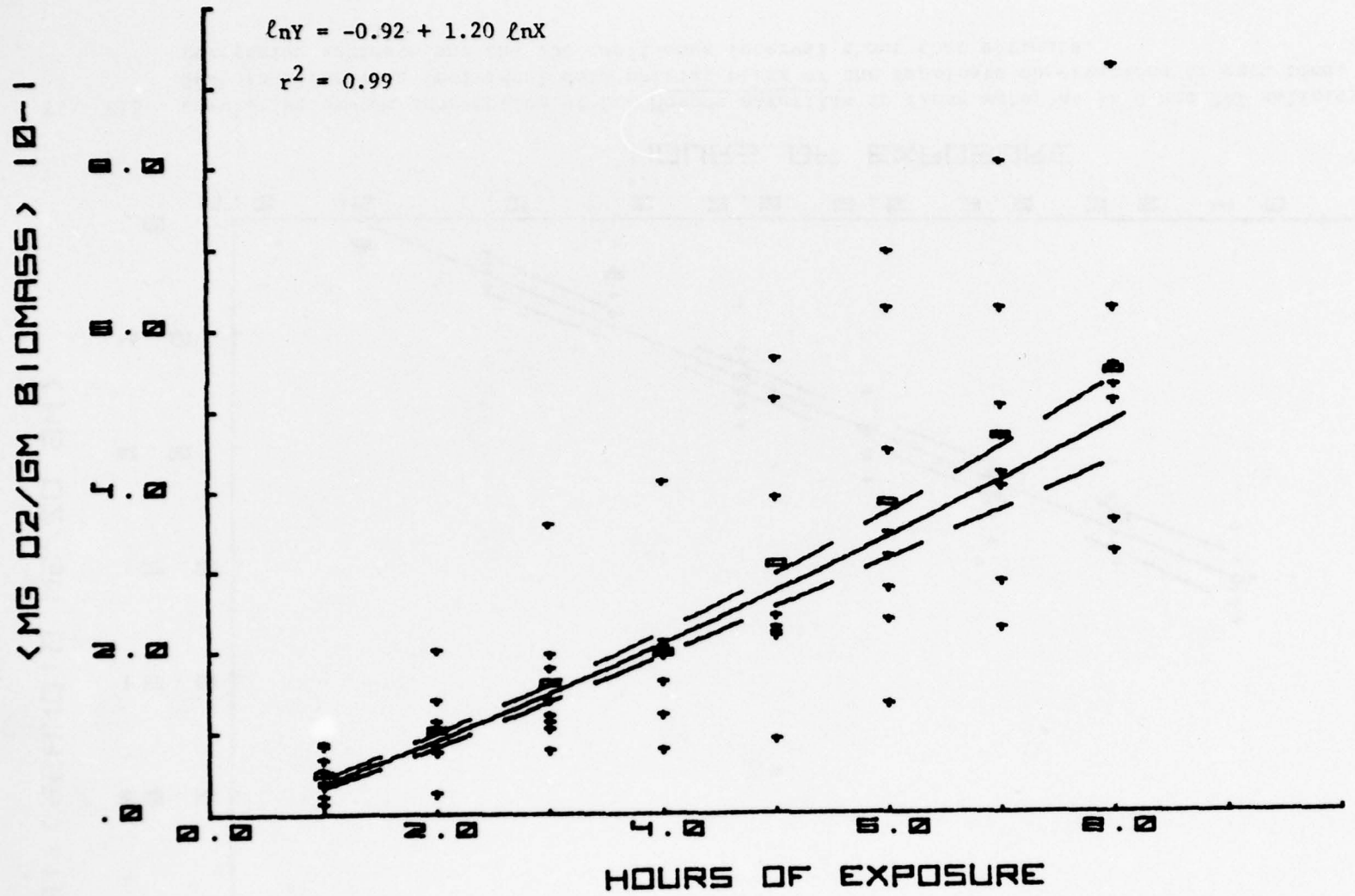

Fig. E12. Cumulative oxygen consumption of 4cm Crangon nigricauda in a starting suspended bentonite concentration of 10 gm/l at 18°C and 28% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

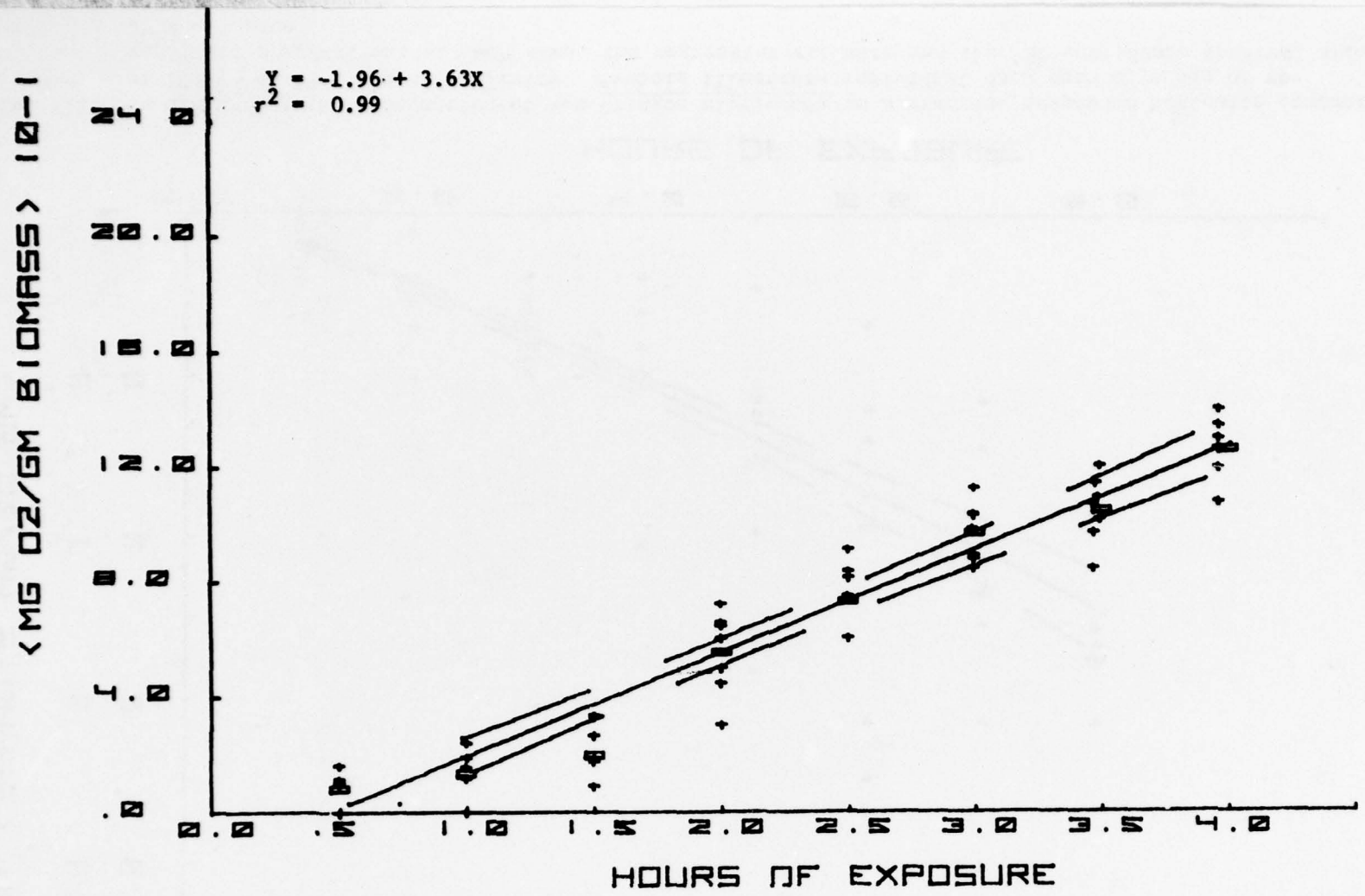

Fig. E13. Cumulative oxygen consumption of 6cm Morone saxatilis in clear water at 18°C and 24% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

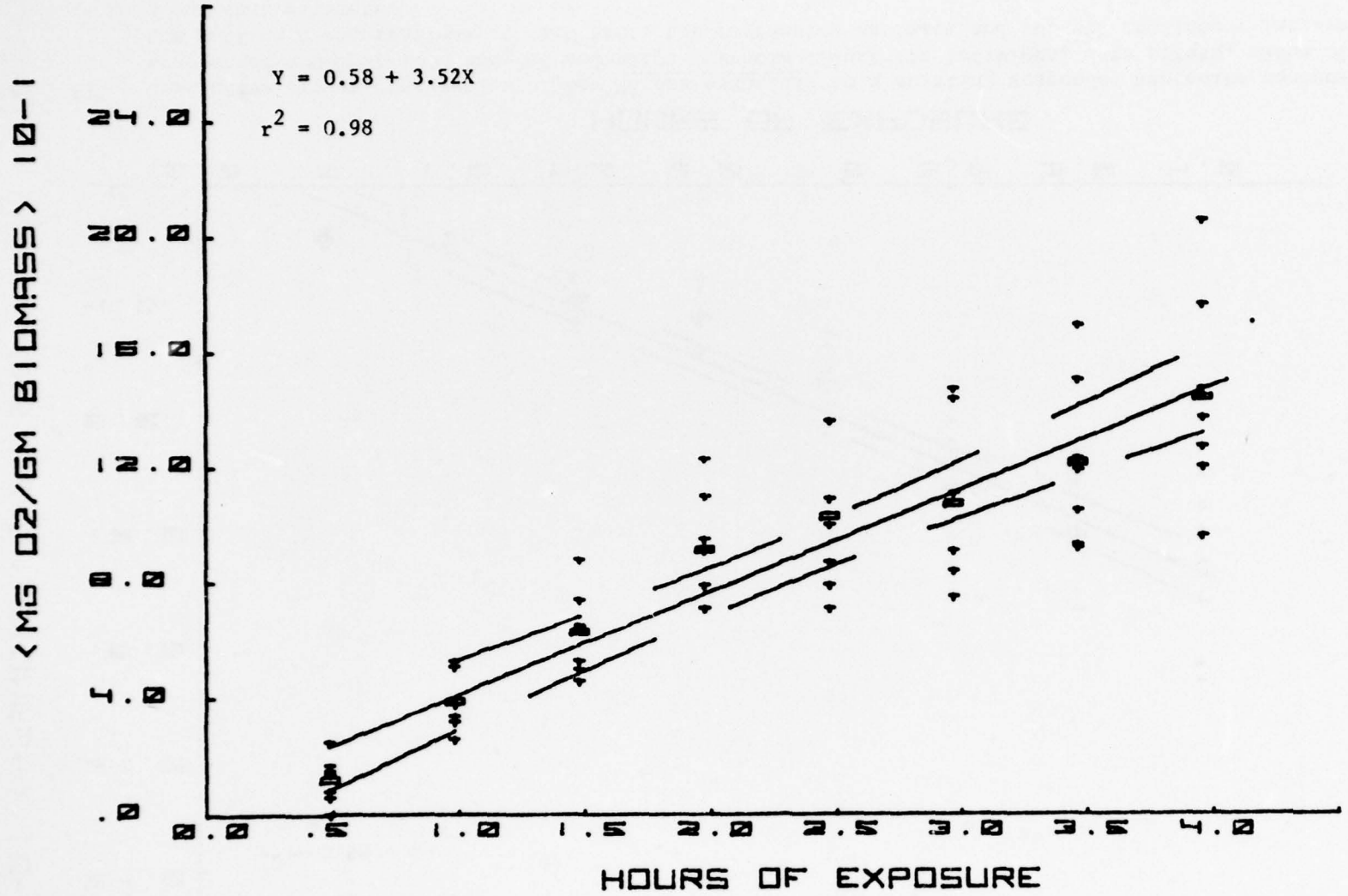

Fig. E14. *Cumulative oxygen consumption of 6cm* Morone saxatilis *in a starting suspended bentonite concentration of 2 gm/l at 18°C and 24% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.*

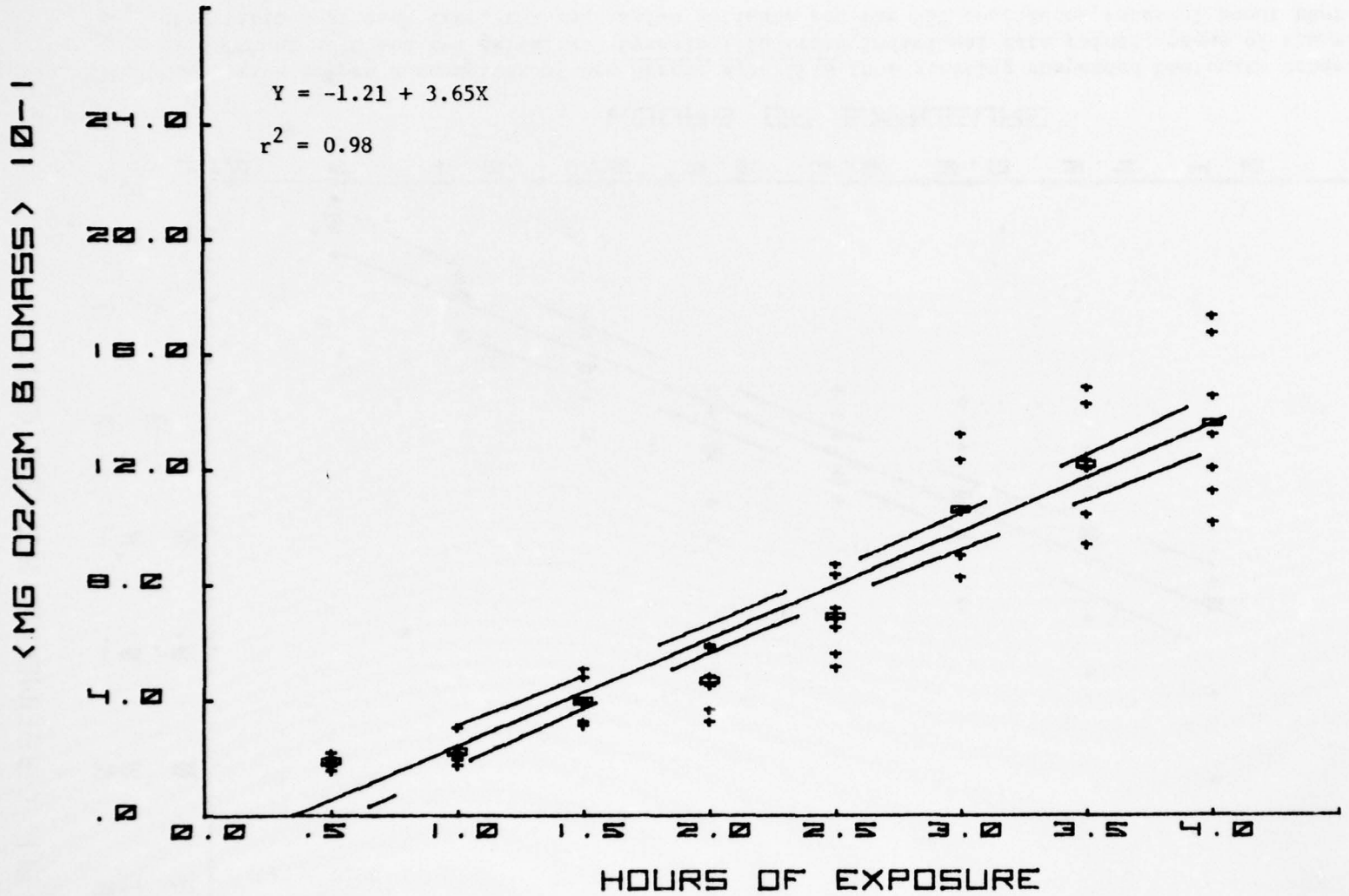

Fig. E15. Cumulative oxygen consumption of 6cm Morone saxatilis in a starting suspended bentonite concentration of 4 gm/l at 18°C and 24% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

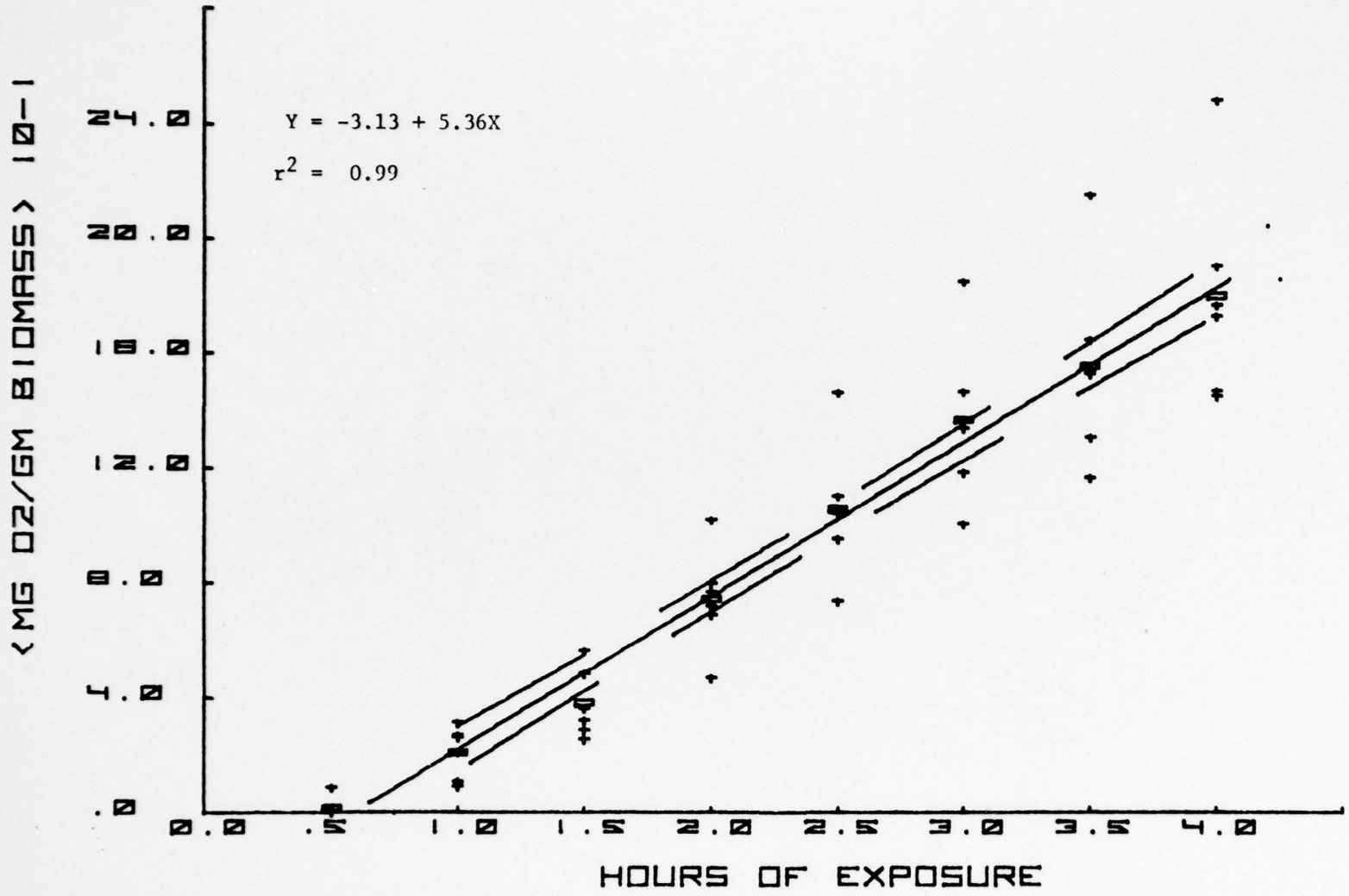

Fig. E16. Cumulative oxygen consumption of 6cm Morone saxatilis in a starting suspended bentonite concentration of 10 gm/l at 18°C and 24% salinity. Symbols illustrate individual data points, means of the replicate observations at each time, the regression estimate and the 95% confidence interval about that estimate.

APPENDIX F

Animal Collection and Holding Data and Experimental Conditions for Sediment Deposition Experiments

APPENDIX F. BURIAL MORTALITY EXPERIMENTS
COLLECTION AND HOLDING DATA

TABLE FI. List of species tested, date, method and site of collection and the salinity and temperature from which the animals were collected. Also presented is the length of time each species was maintained in the laboratory before testing began, the salinity at which they were held, the initial temperature and the final temperature to which each was acclimated for testing. Temperature was changed from the initial to the final at a rate of 2°C every 2 days.

	COLLECTION DATA					HOLDING DATA			
Species	date (1974)	method	site	sal. ppt	temp. °C	days in lab	sal. ppt	init. temp. °C	final temp. °C
Morone saxatilis		HATCHERY FISH					24-28	18	16
Cymatogaster aggregata	early Sept.	otter trawl	San Bruno Shoal area	27-30	18-20	6	24-28	18	16
Crangon nigricauda	late Sept.	otter trawl	San Bruno Shoal area	27-30	18-20	30	26-30	18	14
Synidotea laticauda	late Sept.	dipnet	Central S.F. Bay	25-27	18-20	26	26-30	18	14
Mytilus edulis	early Oct.	hand	Central S.F. Bay	27-30	18-20	20	26-30	18	14

APPENDIX F. BURIAL MORTALITY EXPERIMENTS
EXPERIMENTAL CONDITIONS

TABLE FII. List of species tested, depths of deposited bentonite and salinity and temperature in each aquarium. Species listed together were tested simultaneously in the same aquaria.

Species	0 cm		2 cm		4 cm		6 cm		8 cm	
	sal. o/oo	temp. °C	sal. o/oo	temp. °C	sal. o/oo	temp. °C	sal. o/oo	temp. °C	sal. o/oo	temp. °C
Morone saxatilis	27	14	27	14	27	14	27	14	27	14
Cymatogaster aggregata	27	14	27	14	27	14	27	14	27	14
Crangon nigricauda	30	12	30	12	30	12	30	12	30	12
Synidotea laticauda	30	12	30	12	30	12	30	12	30	12
Mytilus edulis	30	12	30	12	30	12	30	12	30	12