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FIELD VERIFICATION PROGRAM (AQUATIC DISPOSAL)

TECHNICAL REPORT D-87-7

EFFECTS OF BLACK ROCK HARBOR DREDGED MATERIAL ON THE SCOPE FOR GROWTH OF THE BLUE MUSSEL, *MYTILUS EDULIS*, AFTER LABORATORY AND FIELD EXPOSURES

by

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Final Report

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and

US Environmental Protection Agency
Washington, DC 20460

Monitored by Environmental Laboratory
US Army Engineer Waterways Experiment Station
PO Box 631, Vicksburg, Mississippi 39180-0631

US Army Corps of Engineers

SUBJECT: Transmittal of Field Verification Program Technical Report Entitled "Effects of Black Robk Harbor Dredged Material on the Scope for Growth of the Blue Mussel, *Mytilus Edulis*, after Laboratory and Field Exposures"

TO: All Report Recipients

1. This is one in a series of scientific reports documenting the findings of studies conducted under the Interagency Field Verification of Testing and Predictive Methodologies for Dredged Material Disposal Alternatives (referred to as the Field Verification Program or FVP). This program is a comprehensive evaluation of environmental effects of dredged material disposal under conditions of upland and aquatic disposal and wetland creation.

2. The FVP originated out of the mutual need of both the Corps of Engineers (Corps) and the Environmental Protection Agency (EPA) to continually improve the technical basis for carrying out their shared regulatory missions. The program is an expansion of studies proposed by EPA to the US Army Engineer Division, New England (NED), in support of its regulatory and dredging missions related to dredged material disposal into Long Island Sound. Discussions among the Corps' Waterways Experiment Station (WES), NED, and the EPA Environmental Research Laboratory (ERLN) in Narragansett, RI, made it clear that a dredging project at Black Rock Harbor in Bridgeport, CT, presented a unique opportunity for simultaneous evaluation of aquatic disposal, upland disposal, and wetland creation using the same dredged material. Evaluations were to be based on technology existing within the two agencies or developed during the six-year life of the program.

3. The program is generic in nature and will provide techniques and interpretive approaches applicable to evaluation of many dredging and disposal operations. Consequently, while the studies will provide detailed site-specific information on disposal of material dredged from Black Rock Harbor, they will also have great national significance for the Corps and EPA.

4. The FVP is designed to meet both Agencies' needs to document the effects of disposal under various conditions, provide verification of the predictive accuracy of evaluative techniques now in use, and provide a basis for determining the degree to which biological response is correlated with bioaccumulation of key contaminants in the species under study. The latter is an important aid in interpreting potential biological consequences of bioaccumulation. The program also meets EPA mission needs by providing an opportunity to document the application of the generic predictive hazard-assessment research strategy applicable to all wastes disposed in the aquatic environment. Therefore, the ERLN initiated exposure-assessment studies at the aquatic disposal site. The Corps-sponsored studies on environmental consequences of aquatic disposal will provide the effects assessment necessary to complement the EPA-sponsored exposure assessment, thereby allowing ERLN to develop and apply a hazard-assessment strategy. While not part of the Corps-funded FVP, the EPA exposure-assessment studies will complement the Corps' work, and together the Corps and the EPA studies will satisfy the needs of both agencies.

SUBJECT: Transmittal of Field Verification Program Technical Report Entitled "Effects of Black Robk Harbor Dredged Material on the Scope for Growth of the Blue Mussel, *Mytilus Edulis*, after Laboratory and Field Exposures"

5. In recognition of the potential national significance, the Office, Chief of Engineers, approved and funded the studies in January 1982. The work is managed through the Environmental Laboratory's Environmental Effects of Dredging Programs at WES. Studies of the effects of upland disposal and wetland creation were conducted by WES, and studies of aquatic disposal were carried out by the ERLN, applying techniques worked out at the laboratory for evaluating sublethal effects of contaminants on aquatic organisms. These studies were funded by the Corps while salary, support facilities, etc., were provided by EPA. The EPA funding to support the exposure-assessment studies followed in 1983; the exposure-assessment studies are managed and conducted by ERLN.

6. The Corps and EPA are pleased at the opportunity to conduct cooperative research and believe that the value in practical implementation and improvement of environmental regulations of dredged material disposal will be considerable. The studies conducted under this program are scientific in nature and are published in the scientific literature as appropriate and in a series of Corps technical reports. The EPA will publish findings of the exposure-assessment studies in the scientific literature and in EPA report series. The FVP will provide the scientific basis upon which regulatory recommendations will be made and upon which changes in regulatory implementation, and perhaps regulations themselves, will be based. However, the documents produced by the program do not in themselves constitute regulatory guidance from either agency. Regulatory guidance will be provided under separate authority after appropriate technical and administrative assessment of the overall findings of the entire program.

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A study was conducted to investigate residue-effect relationships between tissue residue concentrations and the scope for growth of the blue mussel, *Mytilus edulis*, after exposure in the laboratory and the field to dredged material from Black Rock Harbor (BRH), Bridgeport, Conn. A second objective included field verification of the laboratory results.

A laboratory system was used to provide a constant exposure concentration ranging from 0 to 10 mg/ℓ of suspended BRH sediment. Residue concentrations in mussels, particularly stable compounds such as polychlorinated biphenyls, were found to be closely related to exposure concentration. Scope for growth, clearance rates, and shell growth measurements were inversely related to BRH exposure and subsequent tissue residues, with concentrations as low as 1.5 mg/ℓ of BRH material causing negative biological effects.

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19. ABSTRACT (Continued).

In the field, mussels were placed along a transect from the center of the disposal mound to a clean area distant from the disposal mound. Exposure estimates indicated that the maximum concentration of BRH material occurred during the disposal operation, after which both exposure and tissue residue concentrations decreased dramatically. Of the measurements made at the four field stations during the course of the study, a reduction in the scope for growth of mussels, attributable to BRH material, was observed only once. The estimated concentration of BRH suspended material (0.7 to 0.2 mg/ℓ) during that collection, 8 weeks postdisposal, was very close tc the lowest concentration affecting the scope for growth in the laboratory experiments (1.5 mg/ℓ).

PREFACE

This report describes work performed by the US Environmental Protection Agency (EPA), Environmental Research Laboratory, Narragansett, R. I. (ERLN), as part of the Interagency Field Verification of Testing and Predictive Methodologies for Dredged Material Disposal Alternatives Program (Field Verification Program (FVP)). The FVP was sponsored by the Office, Chief of Engineers (OCE), and was assigned to the US Army Engineer Waterways Experiment Station (WES), Vicksburg, Miss. The objective of this interagency program was to field verify existing predictive techniques for evaluating the environmental consequences of dredged material disposal under aquatic, intertidal, and upland conditions. The aquatic portion of the FVP was conducted by ERLN, with the wetland and upland portions conducted by WES.

The principal investigators for this aquatic study and authors of this report were Mr. William G. Nelson and Dr Richard J. Pruell of Science Applications International Corporation (SAIC) and Dr. Donald K. Phelps, Mr. Walter B. Galloway, and Dr. Peter F. Rogerson of ERLN. Laboratory-cultured algae were provided by Mr. Gregory Tracey, SAIC. Technical support for the scope for growth measurements was provided by Mr. William Giles, ERLN. Diving support for the field portion of the study was provided by Messrs. Bruce Reynolds and Norman Rubenstein, ERLN, and Mr. Tracey, SAIC.

Analytical chemistry support was provided by Dr. Gerald Hoffman, Mr. Richard Lapan, Mr. Curtis Norwood, and Mr. Frank Osterman, ERLN; Mr. Richard McKinney, Mr. Warren Boothman, Ms. Adria Elskus, Ms. Eileen McFadden, Mr. Lawrence LeBlanc, Mr. Robert Bowen, and Ms. Sharon Pavignano, SAIC; and Ms. Kathleen Schweitzer, University of Rhode Island.

Mses. Joan E. Seites, Barbara G. Gardner, and Colette J. Brown, Computer Sciences Corporation (CSC), provided word processing support in preparation of the draft report. In addition, assistance in statistical analysis was provided by Dr. James Heltshe, CSC, and Dr. Clifford H. Katz, SAIC. Critical reviews of this report were provided by Dr. Katz, SAIC, and Drs. John H. Gentile and Gerald G. Pesch, ERLN. Technical reviews by WES personnel were also provided.

The OCE Technical Monitors were Drs. John Hall, Robert J. Pierce, and William L. Klesch. The EPA Technical Director for the FVP was

Dr. John Gentile; Technical Coordinators were Mr. Walter Galloway and Dr. Gerald Pesch; and Project Manager was Mr. Allan Beck.

The study was conducted under the direct WES management of Drs. Thomas M. Dillon and Richard Peddicord and under the general management of Dr. C. Richard Lee, Chief, Contaminant Mobility and Criteria Group; Mr. Donald L. Robey, Chief, Ecosystem Research and Simulation Division; and Dr. John Harrison, Chief, Environmental Laboratory. The Environmental Effects of Dredging Programs Manager was Dr. Robert M. Engler, with Mr. Robert L. Lazor, FVP Coordinator. Dr. Thomas D. Wright was the WES Technical Coordinator for the FVP reports. The report was edited by Ms. Jessica S. Ruff of the WES Information Products Division.

COL Dwayne G. Lee, CE, was the Commander and Director of WES. Dr. Robert W. Whalin was Technical Director.

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CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF FIGURES (Continued)

LIST OF FIGURES (Continued)

EFFECTS OF BLACK ROCK HARBOR DREDGED MATERIAL ON THE SCOPE FOR GROWTH OF THE BLUE MUSSEL, *MYTILUS EDULIS*, AFTER LABORATORY AND FIELD EXPOSURES

PART I: INTRODUCTION

Background

1. The Marine Protection, Research, and Sanctuaries Act (Public Law 92-532) was passed by Congress in 1972. This law states that it is the policy of the United States to regulate disposal of all types of materials into ocean waters and to prevent or strictly limit disposal of any material that would adversely affect human health, welfare, the marine environment, or ecological systems. The implementation of this law, through the issuance of permits as defined in the final regulations and criteria, is shared jointly by the US Environmental Protection Agency (USEPA) and the US Army Corps of Engineers (CE).

2. In 1977, the CE and the USEPA prepared technical guidance for the implementation of the final ocean dumping regulations in the form of a manual entitled "Ecological Evaluation of Proposed Discharge of Dredged Material into Ocean Waters" (USEPA/CE 1977). This manual specified which test procedures were to be followed in collecting information to be used in making a disposal decision. Among the procedures were those for: (a) chemically characterizing the proposed dredged material; (b) determining the acute toxicity of liquid, suspended particulate, and solid phases; (c) estimating the potential contaminant bioaccumulation; and (d) describing the initial mixing during disposal. These methods have been used for determining the suitability of dredged material for open-water disposal. The procedures in this manual represented the technical state of the art at that time and were never intended to be inflexible methodologies. The recommended test methods were chosen to provide technical information that was consistent with the criteria specified in the regulations. However, use of the manual in the permit process has identified conceptual and technical limitations with the recommended test methods (Gentile and Scott 1986).

3. To meet this critical need, the Interagency Field Verification of Testing and Predictive Methodologies for Dredged Material Disposal Alternatives Program or the Field Verification Program (FVP) was authorized in 1982. This 6-year program was sponsored by the Office, Chief of Engineers, and was assigned to the US Army Engineer Waterways Experiment Station (WES), Vicksburg, Miss. The objective of this interagency program was to field verify existing test methodologies for predicting the environmental consequences of dredged material disposal under aquatic, intertidal, and upland conditions. The aquatic portion of the FVP was conducted by the USEPA Environmental Research Laboratory, Narragansett, R. I. (ERLN). The wetland and upland portions, conducted by WES, are reported in separate documentation.

4. The USEPA ERLN was responsible for conducting research on the aquatic option for disposal of dredged material. There were three research objectives for this portion of the program. The first was to demonstrate the applicability of existing test methods to detect and measure effects of dredged material, and to determine the degree of variability and reproducibility inherent in the testing procedure. This phase of the program (Laboratory Documentation) is complete and the results are published in a series of technical reports. This information provides insight into how the various methods function, their sources of variability, their respective and relative sensitivities to the specific dredged material being tested, and the degree of confidence that can be placed on the data derived from the application of the methods.

5. The second objective was to field verify the laboratory responses by measuring the same response under both laboratory and field exposures. A basic and often implicit assumption is that results derived from laboratory test methods are directly applicable in the field. While this assumption is intuitive, there are no supporting data from studies on complex wastes in the marine environment. The study reported herein offers a unique opportunity to test this basic assumption.

6. The third objective was to determine the degree of correlation of tissue residues resulting from bioaccumulation of dredged material contaminants with biological responses from laboratory and field exposure to dredged material. However, this study was not designed to address cause-effect relationships, and the multicontaminant nature of the dredged material precludes any such assumptions.

Project Description

7. The aquatic disposal portion of the FVP was a site- and waste-specific case study that applied the concepts and principles of risk assessment. The disposal site for the FVP was an historical site known as the Central Long Island Sound (CLIS) disposal site (1.8 by 3.7 km) located approximately 15 km southeast of New Haven, Conn. (Figure 1). The sedimentology at

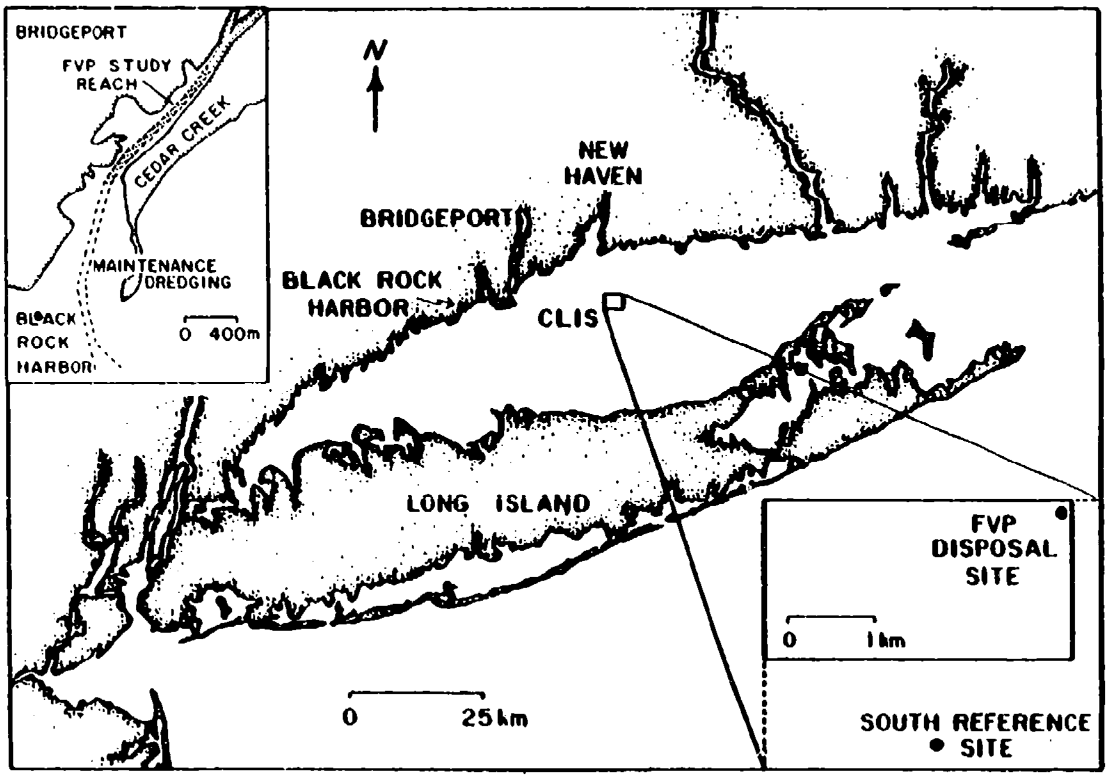


Figure 1. Central Long Island Sound disposal site and Black Rock Harbor dredge site

the disposal and reference sites is primarily silt-clay, with a mean grain size of 0.013 mm. Thermal stratification occurs from April to September, and during this period bottom salinity is slightly higher than that of the surface. Tidal currents typically dominate the near-bottom water in an east-west direction. The net bottom drift is to the northwest at 0.5 cm/sec. Suspended sediment concentrations average 10 mg/ℓ, with storm-induced values to 30 mg/ℓ. The baseline community data revealed a homogeneous, mature infaunal community dominated by the polychaete *Nephtys incisa* and the bivalve molluscs *Nucula proxima* and *Yoldia limatula*.

8. The FVP disposal site was selected within the CLIS so as to minimize contamination from other sources, including relic disposal operations or ongoing disposal activities occurring during the study period. This was necessary to ensure a point source of contamination. The uniformity of physical, chemical, and biological properties of the disposal site prior to disposal allowed detection of changes in these properties due to the disposal of the dredged material. Finally, the stations used to study the biological effects in this study were selected along the primary axis of current flow to represent a gradient of potential exposure for the biota (Figure 2).

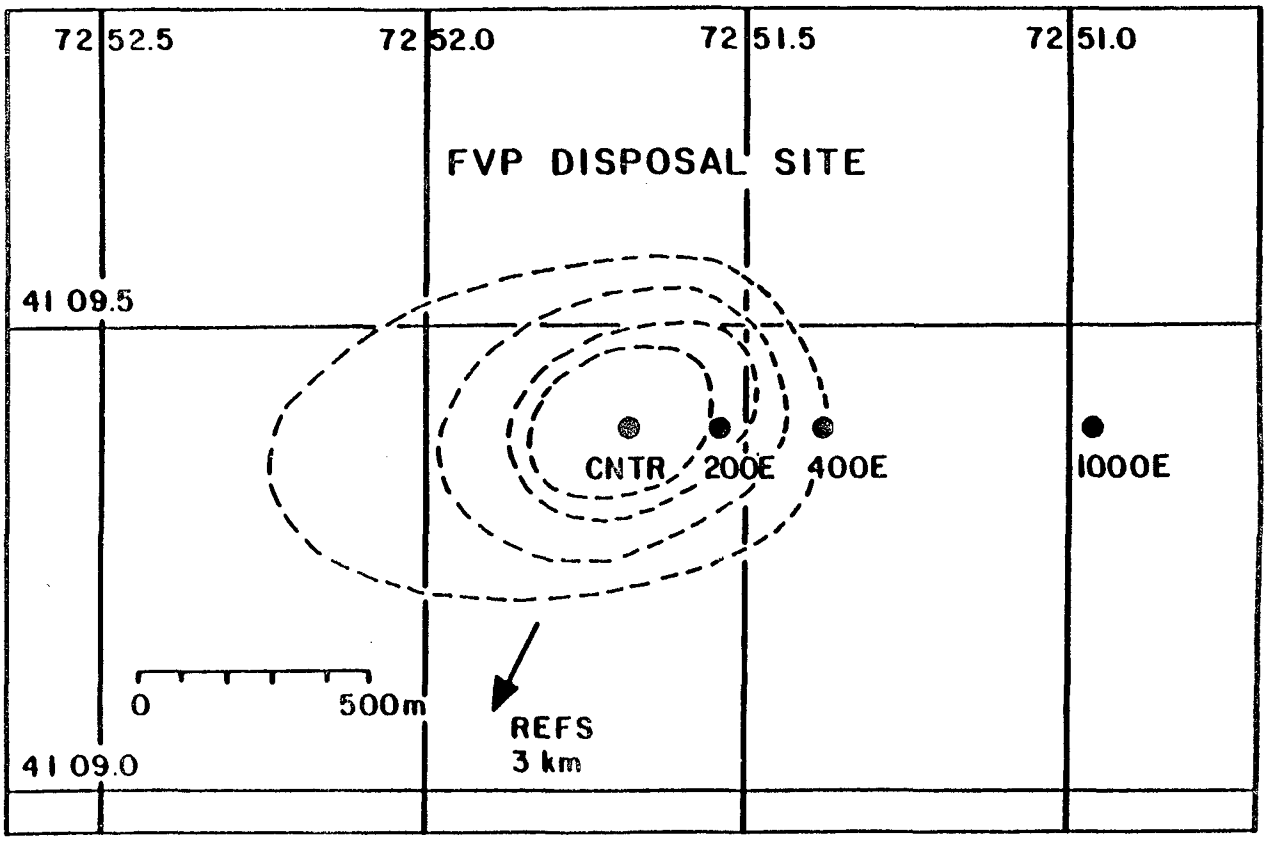


Figure 2. FVP sampling stations

9. The spatial scale of this study was near-field and limited to the immediate vicinity of the disposal site. A primary assumption was that the mound of dredged material constitutes a point source of contamination. The temporal scale for the study was 4 years, which included a year of predisposal data collection to define seasonal patterns in the physical, chemical, and biological variables and 3 years of postdisposal data collection to address the objectives of the program and to evaluate the long-term impacts of the disposal operation on the surrounding benthic communities.

10. The dredging site was Black Rock Harbor (BRH), located in Bridgeport, Conn., where maintenance dredging provided a channel 46 m wide and 5.2 m deep at mean low water (Figure 1). Approximately 55,000 m^3 of material were dredged during April and May 1983 and disposed in 20 m of water in the northeastern corner of the CLIS disposal site.

11. The dredged material from BRH contained substantial concentrations of both organic and inorganic contaminants (Rogerson, Schimmel, and Hoffman 1985). Polychlorinated biphenyls (PCBs) were present in the dredged material at a concentration of 6,800 ng/g, and polynuclear aromatic hydrocarbons (PAHs) with molecular weights between 166 and 302 were present at concentrations ranging from 1,000 to 12,000 ng/g. Alkyl homologs of the PAHs were also present in the dredged material at concentrations between 1,000 and 13,000 ng/g. Inorganic contaminants of toxicological importance present in the dredged material include copper (2,380 μg/g), chromium (1,430 μg/g), zinc (1,200 μg/g), lead (380 μg/g), nickel (140 μg/g), cadmium (23 μg/g), and mercury (1.7 μg/g).

Project Scope

12. The FVP was unique among marine research studies for several reasons. The program objectives were directly focused on addressing specific limitations in the methodologies and interpretive framework of the current regulatory process. Among the program strengths were: the development and evaluation of a suite of biological endpoints that used the same material; the biological tests represented different levels of biological organization; the tests were conducted under both laboratory and field exposure conditions; tissue residues were examined concurrently with measurements of biological effects; the duration of the study was adequate to evaluate the use of community responses as a benchmark against which other biological responses could be compared; and the project was a site- and waste-specific case study for the application and evaluation of the components of a risk assessment, including the development of methodologies for predicting and measuring field exposures in the water column and benthic compartments. Limitations of this study were: only one dredged material was evaluated, which constrained certain types of comparisons; the size of the study put limits on the extent to which any given objective could be examined; and the resources allocated to determine field

exposures were insufficient. The last constraint was particularly important because the laboratory-field comparisons and the risk assessment process both required accurate predictions of environmental exposures.

Laboratory-Field Comparisons

13. The field verification of laboratory test methods was designed to compare the exposure-response relationships measured in both the laboratory and the field. Exposure for the purposes of this discussion includes the total dredged material with all of its contaminants. Specific contaminants are used as "tracers" to verify the exposure environment, which is described in terms of BRH dredged material, and to illustrate exposure-response relationships between the laboratory and field. The specific contaminants are a subset of a comprehensive suite of chemicals analyzed in this study and were selected based upon their environmental chemistry and statistical representativeness. The use of specific contaminants in no way implies a cause-and-effect relationship between contaminant and response.

14. Exposure in open marine systems is characterized by highly dynamic temporal and spatial conditions and cannot be completely replicated in laboratory systems. Consequently, the approach chosen for this program was to develop laboratory exposure-response data using only general field exposure information. The most appropriate statistical analyses for laboratory-field comparisons are observational where all the variables from the laboratory and field are used to identify similarities and differences, independent of any limiting assumptions.

Residue-Effects Relationships

15. Determining the relationship between contaminant tissue residues resulting from bioaccumulation and the biological responses measured is a principal objective of this program. Such relationships do not in any way imply cause and effect, but rather seek to determine the statistical relationship between an effect and any associated residues. The approach used is to determine specific contaminant residues in the tissues of the organisms as the result of exposure to the whole dredged material in both the laboratory and the field. These residues are determined at the same time that biological

responses are being measured. Residue-effect relationships will be described and interpreted for both laboratory and field exposures.

Scope for Growth

16. The scope for growth (SFG) index (Warren and Davis 1967) is a measure of the energy available to an organism for production, both somatic and reproductive, after accounting for routine metabolic costs. The SFG value represents the instantaneous assessment of energy balance in an organism for that set of environmental conditions under which it is measured.

17. A very important point pertinent to the interpretation of SFG measurements is that this index may be used to test several hypotheses. The present study investigated the hypothesis that exposure to different environmental conditions (concentrations of BRH dredged material) may result in lasting physiological effects in an organism, even after its removal from those exposure conditions. Consistent with this hypothesis, the SFG of mussels was measured under standardized conditions, after separate laboratory and field exposures. These measurements were made to test for relative differences between stations (field) or between exposure concentrations (laboratory). SFG differences in this instance were interpreted as being caused by the respective environmental conditions to which the organisms were exposed. This is true since, under standardized conditions, mussels of similar physiological condition should exhibit similar SFG responses. This approach was used because one of the goals of this study was to compare the laboratory and field SFG results. Measurement of SFG under separate laboratory and field conditions would not allow this comparison.

18. There are sufficient historical data to indicate that the use of the SFG index with the blue mussel, *Mytilus edulis*, might be useful in assessing the biological impact of disposed dredged material in the marine environment. The SFG index proved useful as a response parameter for measuring physiological effects of BRH dredged material on *M. edulis* in the laboratory (Nelson, Black, and Phelps 1985). Stickle et al. (1985) reported an inverse relationship between the SFG of mussels and exposure to increasing concentrations of the water-soluble fraction of crude oil. Widdows et al. (1982) also reported a dose-response effect between SFG and aromatic petroleum hydrocarbon exposure concentrations in mussels. In the field, Widdows, Phelps, and

Galloway (1981) reported a decrease in the SFG of mussels in response to increasing levels of pollution in Narragansett Bay.

19. The objectives of this portion of the FVP with respect to the SFG index were to: (a) determine whether there was a relationship between contaminant residues in tissues and subsequent biological effects measured in both the laboratory and the field, and (b) determine whether the responses measured in the laboratory exposures were comparable to the responses measured in field organisms for similar exposures. The evaluation of these objectives will form the basis of this field verification report.

PART II: MATERIALS AND METHODS

Chemical Methods

Analytical methods

20. The analytical methods used in this study are presented here in summary form. More detailed descriptions of the analytical methods are available in Lake, Hoffman, and Schimmel (1985). Most of these methods represent extensive modifications of USEPA standard methods developed for freshwater and wastewater samples. It was necessary to modify these methods to analyze the types of matrices in this study. These methods were intercalibrated to ensure the quality of the data.

Organic sample preparation

21. Samples of sediment, suspended particulates, and organisms were extracted by multiple additions of increasingly less polar organic solvents using a tissue homogenizer. These mixtures were separated by centrifugation between additions; polar solvents were removed by partitioning against water; and the extracts were desulfured with activated copper powder when required. The extracts were then passed through a precolumn containing activated silica gel. Samples of both filtered and unfiltered seawater were solvent extracted in separatory funnels and the extracts saved. Foam plugs containing the dissolved organic contaminants from water samples were extracted with organic solvents. All of the above extracts were subjected to column chromatography on deactivated silica gel to separate analytical fractions and were volume reduced carefully prior to analysis.

Organic analysis

22. Electron capture gas chromatographic analyses for PCBs were conducted on a Hewlett-Packard 5840 gas chromatograph equipped with a 30-m DB-5 fused silica column. Samples were quantified against an Aroclor 1254 (A1254) standard because the distribution of PCB congeners in the dredged material closely matched that distribution, as did the distribution in organisms at steady state.

23. Gas chromatograph/mass spectrometric analyses were conducted with a Finnigan Model 4500 also equipped with a 30-m DB-5 fused silica capillary column. The mass spectrometer was operated through a standard Incos data system and was tuned at all times to meet USEPA quality assurance specifications.

24. All instruments were calibrated daily with the appropriate standards. The concentrations of the standards used were chosen to approximate those of the contaminants of interest, and periodic linearity checks were made to ensure the proper performance of each system. When standards were not available, response factors were calculated using mean responses of comparable standards. Blanks were carried through the procedure with each set of samples, and reference tissue homogenate was analyzed with every 12 to 15 tissue samples.

<u>Organic data compression</u>

25. As stated above, PCBs were quantified as A1254 because the sample patterns closely resembled that profile. This allowed a convenient way of reporting these data without treating the voluminous data that would have resulted from measuring some 55 congener peaks by electron capture detector. Likewise, a method was sought to summarize the PAH data. Appendix A lists the 35 individual PAH parent and alkyl homolog compounds and groups of compounds measured in this study. Although useful, this only reduced the data to 14 PAH variables, which was not sufficient. Since the distribution of PAHs differed greatly in both quantity and distribution between Long Island Sound and the BRH dredged material, statistics were sought which would retain significant quantitative and qualitative information. The quantitative statistic chosen was the simple SUM of all measured PAHs, and a qualitative descriptor was chosen by analogy with the center of mass concept from elementary physics and called a centroid (CENT):

$$\mathrm{SUM} = \Sigma[C(i)] \tag{1}$$

$$\mathrm{CENT} = \frac{\Sigma[C(i) * MW(i)]}{\mathrm{SUM}} \tag{2}$$

where

$C(i)$ = concentration of i^{th} PAH from molecular weight 166 through 302, including both parent and alkyl homologs

$MW(i)$ = molecular weight of i^{th} PAH from 166 through 302, including both parent and alkyl homologs

In this case, CENT describes the "center of mass" of the PAH distribution, and is in units of molecular weight. It is the concentration-weighted average molecular weight of any particular PAH distribution. Using this statistic, one is able to readily distinguish two different sources of PAH distributions, one with predominantly heavy molecular weight pyrogenic compounds, and one with more lighter molecular weight petrogenic compounds. These distributions are typically found at the reference station in Long Island Sound (REFS) and BRH, respectively. A major value to this statistic is that it enables one to readily distinguish these two sources when their concentrations are nearly equal. The formulas for calculating these, and 178 alkyl homologs, are shown in Appendix A. Because distributions of both parents and homologs were measured, SUMs and CENTs of both parents and homologs were calculated as well. These were defined as PSUM, PCENT, HSUM, and HCENT. By definition,

$$SUM = PSUM + HSUM \tag{3}$$

and

$$CENT = \frac{(PSUM * PCENT + HSUM * HCENT)}{SUM} \tag{4}$$

It should be noted that dibenzothiophene and its alkyl homologs are not included in these calculations because they are not PAHs.

Inorganic sample preparation

26. Sediment was prepared for inorganic analysis by elution at room temperature with 2N HNO_3. The samples were filtered through Whatman No. 2 filter paper. Organisms were totally digested in concentrated HNO_3 at 60° C and filtered through Whatman No. 2 filter paper.

27. Cadmium and copper were concentrated and separated from both the unfiltered and filtered seawater fractions by coprecipitation (Boyle and Edmond 1975). The remaining metals (chromium, iron, manganese, nickel, lead, and zinc) were analyzed by heated graphite atomization atomic absorption (HGA-AA) via direct injection. Samples of suspended particulates on Nucleopore (0.45-μ) filters were eluted with 2N HNO_3 and analyzed by HGA-AA.

Inorganic analysis

28. All flame atomization atomic absorption (FA-AA) was conducted with a Perkin-Elmer (Model 5000) atomic absorption spectrophotometer. All HGA-AA determinations were conducted with Perkin-Elmer Model 5000 or 2100 HGA units coupled to Perkin-Elmer Model 5000 or 603 atomic absorption instruments, respectively. The Model 5000 AA was retrofitted with a Zeeman HGA background correction unit, and the Model 603 was equipped with a D2 arc background correction system.

29. The FA-AA and HGA-AA instrument operating conditions are similar to those described in "Methods for Chemical Analysis of Water and Wastes" (USEPA 1979) and those in the manufacturers' reference manuals. The AA instruments were calibrated each time samples were analyzed for a given element. Sample extracts were analyzed a minimum of twice to determine signal reproducibility. Quality assurance checks, conducted after every 15 samples, were analyzed by the method of standard addition and by analyzing one procedural blank.

Contaminant selection

30. Chemical analyses performed in this study characterize the organic and inorganic constituents in the dredged material; provide information on the laboratory and field exposure environments; provide insight into the processes governing contaminant movement within and between environmental compartments; and determine which contaminants were accumulated by organisms. Historically, bulk sediment analyses have been used to characterize dredged material. More recently, dredged material must be analyzed for USEPA's priority pollutants to determine if hazardous substances are present and, if so, in what concentrations. While both of these approaches were used in this study, neither addresses the issue of bioavailability and the potential for contaminants to bioaccumulate. In this study, bioavailability was determined by examining the types and distributions of contaminants that bioaccumulated in laboratory studies (Rogerson, Schimmel, and Hoffman 1985). Based upon the contaminant profile for the dredged material and residue data, the contaminants selected for detailed analyses throughout the study included PCBs, PAHs, the pesticide ethylan, and four metals.

31. A representative subset of chemicals was selected for discussion throughout the study. The criteria used in selecting this subset included chemical properties, contaminant representativeness and behavior in various compartments, and statistical analyses of the distributions of the complete suite of chemicals analyzed in the program.

32. Multivariate clustering analyses were performed on the chemical data to define groups or clusters of chemicals that behaved in a statistically similar manner. No assumptions were made concerning the behavior, interactions, or dynamics of chemicals between compartments; therefore, each compartment was analyzed separately. Five compartments were identified from field and laboratory data for statistical analysis. Of these, the surficial sediments and the unfiltered, particulate, and dissolved water column

fractions described exposure conditions experienced by infaunal and pelagic organisms. The remaining compartment consisted of tissue residues in *M. edulis*.

33. The data were further partitioned into inorganic and organic analyses. The inorganic analyses generally consisted of 8 variables while the organic analyses contained 61 variables. The clusters of chemicals identified through the statistical analyses agreed well with those contaminants selected based on chemical properties and environmental behavior.

Laboratory Methods

Sediment collection

34. Two sediment types were used to conduct laboratory tests for the field verification studies. The reference sediment (REF) was collected from the South Reference site (REFS) in Long Island Sound (40°7.95' N and 72°52.7' W) by Smith-MacIntyre grab (0.1 m^2), press sieved through a 2-mm sieve, and stored in barrels at 4° C until used. Prior to dredging, contaminated sediment was collected from BRH (41°9' N and 73°13' W) with a gravity box corer (0.1 m^2) to a depth of 1.21 m, thoroughly mixed, press sieved through a 2-mm sieve, and refrigerated (4° C) until used. Details of sediment collection and storage procedures may be found in Rogerson, Schimmel, and Hoffman (1985). In all experiments, sediments were allowed to reach test temperature and were mixed prior to use.

Organism collection and holding

35. Two separate experiments were completed using oxidized REF and BRH sediments. Mussels were collected from the Narragansett Bay reference population (71°24.0' W by 41°29.4' N) with a scallop dredge from a depth of 10 m. Collection information for each experiment is listed in Table 1. The animals

Table 1

Collection Information for the Mussels Used in the Laboratory Experiments

Experiment	Collection Date	Experiment Begun	Temperature °C	Salinity g/kg
1	17 Jan 85	05 Feb 85	2.0	30.0
2	22 Feb 85	12 Mar 85	5.0	30.0

were sorted to obtain a size range of 50- to 55-mm shell length and acclimated in flowing unfiltered Narragansett Bay seawater at a rate of 1° C per day to 15° C.

Suspended sediment dosing system

36. Laboratory studies required the construction of two identical sediment dosing systems to provide either BRH or REF material as suspended sediment simultaneously. Each dosing system (Figure 3) consisted of a

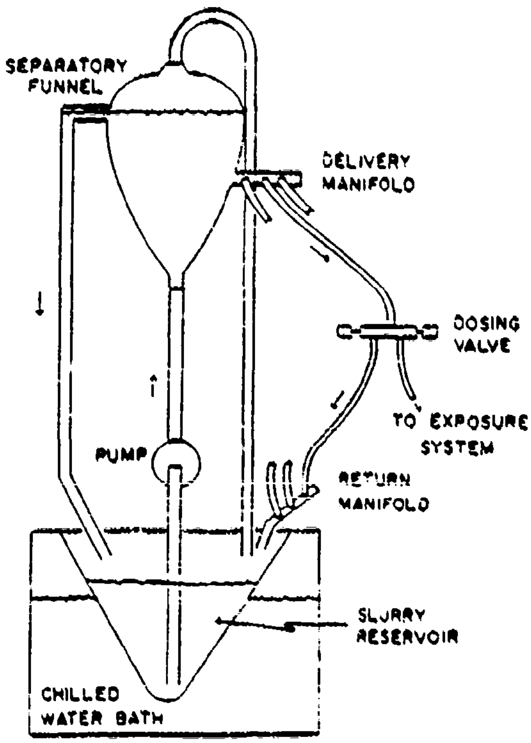


Figure 3. Suspended sediment dosing system

conical-shaped slurry reservoir placed in a chilled fiberglass chamber, a diaphragm pump, a 4-ℓ separatory funnel, and several return loops that directed the particulate slurry through dosing valves. The slurry reservoirs (40 cm in diameter by 55 cm high) contained 40 ℓ of slurry composed of 37.7 ℓ of filtered seawater and 2.3 ℓ of either BRH or REF sediment. The fiberglass chamber (94 by 61 by 79 cm high) was maintained between 4° and 10° C using an externally chilled water source to minimize microbial degradation during the test. Polypropylene pipes (3.8-cm diam) extended to the bottom of the reservoir cones and were connected to pumps (16- to 40-ℓ/min capacity) fitted with Teflon diaphragms. These pumps were used to circulate the slurry while minimizing abrasion which might produce changes in the physical properties (e.g., particle size) of the material.

37. The slurry was pumped up to separatory funnels and returned via an overflow to the reservoir through polypropylene pipes. The separatory funnel provided the constant head pressure needed to circulate the slurry through Teflon tubing to the dosing valves where the slurry was mixed with seawater to provide the desired concentrations for the toxicity tests. Narragansett Bay seawater filtered (to 15 μ) through sand filters was used.

Suspended sediment oxidation system

38. The REF and BRH sediments used in these experiments were oxidized prior to introduction into the dosing system. The objective of this portion of the FVP was to evaluate the relationship between biological endpoints measured in the laboratory and the field. The field collections of sediment indicated rapid oxidation of the surficial BRH sediments on the disposal mound. Because the most likely source of particulate contaminants in the water column was the oxidized surficial sediment, it was decided that laboratory exposures would be conducted with BRH sediment that had been oxidized in a consistent manner.

39. In order to obtain consistent states of oxidation for both REF and BRH sediments, 2 ℓ of sediment were transferred to an inverted polycarbonate carboy and diluted to 19 ℓ with filtered natural seawater at room temperature and aerated for 3 to 4 days (Figure 4). The contents were transferred to the composite dosing system reservoir and diluted to 38 ℓ with natural seawater. Chemical oxygen demand measurements indicated that this time period was sufficient to satisfy the immediate oxygen demand of the sediments.

Exposure system

40. An exposure system was constructed to provide a constant concentration of suspended sediment to mussels in the laboratory. This system consisted of recirculating loops from the suspended sediment dosing system which were connected to a dosing valve at each exposure chamber. The concentration of total suspended particulates was maintained at approximately 12 mg/ml in both the REF and BRH loops. The exposure system was capable of delivering either REF or BRH sediment directly into each mussel exposure chamber via a dosing valve. The combined use of a REF and a BRH dosing valve at an exposure chamber allowed delivery of a mixture of the two sediments. The percent concentrations of BRH and REF sediment varied between treatments; however, a total suspended sediment concentration of approximately 10 mg/ℓ (dry weight) was maintained in all five laboratory exposure treatments. This concentration

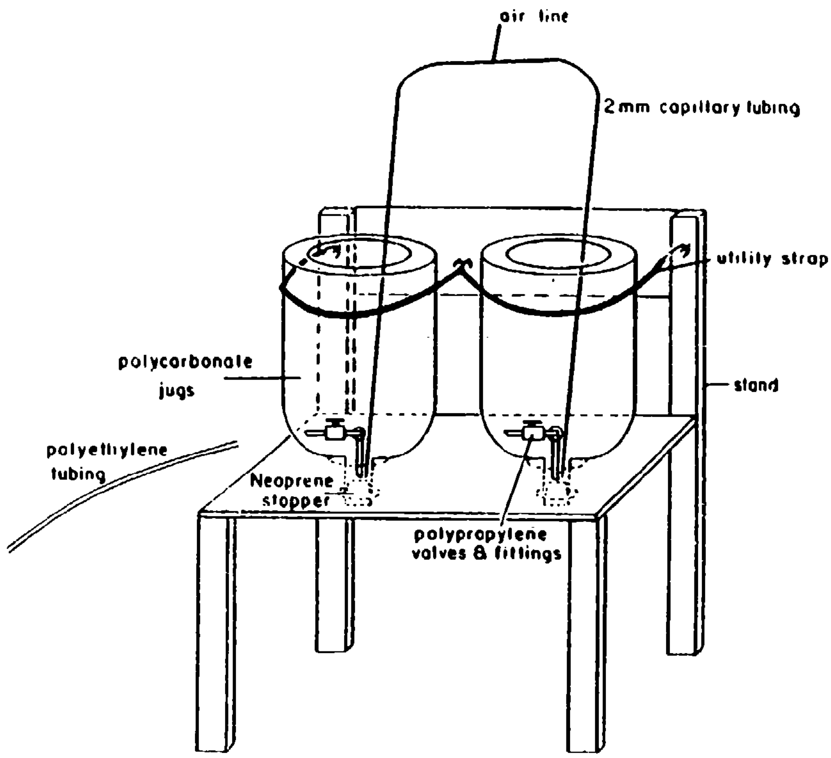
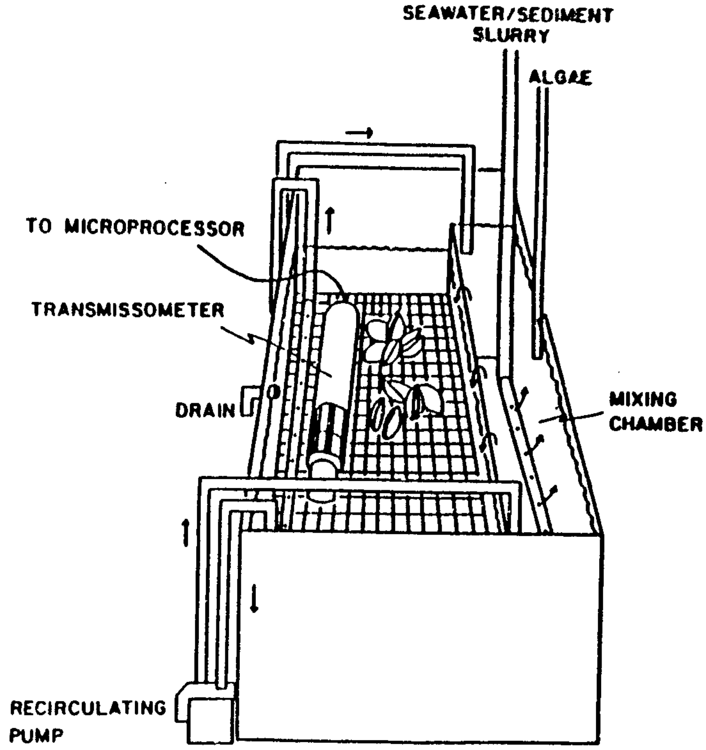


Figure 4. Suspended sediment oxidation system

was chosen because it approximated the background field suspended sediment concentration at the CLIS disposal site.

41. Each mussel exposure chamber was equipped with a transmissometer, an instrument capable of measuring light attenuation due to suspended sediment in the chamber (Figure 5). The dosing valves for each treatment were controlled by a transmissometer-microprocessor feedback loop (Sinnett and Davis 1983). The transmissometer in each chamber was calibrated by regressing suspended sediment concentrations, measured by filtration onto glass fiber filters, with the transmissometer units displayed on a microprocessor. A transmissometer value was calculated that corresponded with the desired suspended sediment concentration of 10 mg/ℓ for each chamber. As the mussels removed suspended sediments, the microprocessor opened dosing valves to deliver additional suspended sediment at 2-min intervals. In this manner, suspended sediment concentrations were maintained at the desired values (±10 percent). The transmissometer circuit was also connected to a strip chart recorder which allowed the operation of the system to be monitored continuously. Each chamber was aerated with three 25- by 2.5-cm air stones to provide sufficient oxygen and to ensure even distribution of suspended particulates (Figure 5).

Figure 5. Laboratory exposure system



42. In addition to the suspended sediment, food in the form of a unicellular alga, *Isochrysis galbana*, was supplied to each exposure chamber. Periodic measurements were made of mussel clearance rates in each chamber to determine the volume of algae required to maintain an algal concentration of 0.5 mg/ℓ. This concentration constituted an adequate maintenance ration for the mussels. Algae were added at 5-min intervals by means of a peristaltic pump. All experiments were conducted at 15° C with filtered seawater that flowed through each experimental chamber at a rate of 0.4 ℓ/min. Each chamber was cleaned every other day.

43. The purpose of the laboratory experiments was to expose *M. edulis* to a range of BRH concentrations that may have been present in CLIS and to assess the biological effect on these organisms. *Mytilus edulis* were exposed for approximately 1-month periods at the CLIS disposal site; therefore, exposures of similar duration, 28 days, were used for the laboratory exposures.

44. At the start of both experiments, 150 mussels were placed into each chamber. *Mytilus edulis* were sampled at time zero to determine initial tissue residue concentrations, and for SFG testing.

45. Experiment 1 consisted of three exposure concentrations: 100-, 50-, and 0-percent BRH suspended sediment. *Mytilus edulis* were removed from each treatment on Day 14 for chemical and biological analysis. Experiment 1

was terminated at Day 14 because adverse biological effects (e.g., reduced filtration rate) were observed in both treatments containing BRH sediment.

46. Experiment 2 was conducted with lower concentrations of BRH suspended sediment. Exposure concentrations of suspended sediment in Experiment 2 were 30-, 10-, and 0-percent BRH. Fifteen organisms were removed on Days 7, 14, 21, and 28 for tissue residue analysis. Whole water chemistry samples were taken within 1 day of organism sampling. Dissolved and particulate water samples were taken within 24 hr of Days 0, 14, and 28. Mussels were sampled on Days 14 and 28 for biological analysis. In addition, a water sample was taken on Day 29 to evaluate the performance of the system without any mussels in the exposure chambers.

47. The operation of the system (dosing valves, flow rates, etc.) was monitored daily. Experiments using the 100- and 0-percent BRH concentrations required only one dosing valve each, while the 50-percent BRH treatment required a REF and BRH valve that delivered equal amounts of suspended material. A strip chart record for each treatment indicated that the dosing valves were operating properly. The 10- and 30-percent BRH treatments also required two dosing valves per treatment; however, the REF and BRH dosing valves delivered different amounts of suspended material. This was accomplished by adjusting the delivery volume of each valve. The mixture of BRH and REF material was checked daily and adjusted if necessary.

SFG procedures

48. Calculation of the SFG index for *M. edulis* required the measurement of four parameters: clearance rate, respiration rate, food absorption efficiency, and ammonia excretion rate. Measurements were completed under standardized conditions: temperature 15° C, salinity 30 ppt, and an algal concentration of 0.5 mg/ℓ. All SFG measurements for a given treatment were completed in the order shown below within 28 hr after termination of the experiment. The detailed methods are described by Nelson, Black, and Phelps (1985); therefore, only a brief summary of each procedure is provided here.

49. Clearance rate. Clearance rate is defined as the volume of water completely cleared of particles >3 μ in some unit of time (Widdows, Fieth, and Worrall 1979). Mussels were placed into individual chambers through which 1 μ filtered seawater flowed at a rate of 75 ml/min. The unicellular alga *T-Iso* was added to the filtered seawater to deliver an incoming cell concentration of approximately 25,000 cells/ml (about 0.5 mg/ℓ) to each chamber. Each

chamber was gently aerated to ensure that complete mixing and no settling of algae occurred. Mussels were allowed to acclimate in the chambers for at least 1 hr prior to any measurements. Incoming and outgoing particle concentrations for each chamber were then measured 3 times at 1-hr intervals with a Coulter Counter (Model TAII).

50. Respiration rate. Respiration rates were determined by isolating each mussel in a glass respirometer vessel fitted with a PO2 electrode. The electrode was connected to a Radiometer oxygen meter (Model PHM71) which was in turn connected to a strip chart recorder. The decline in PO2 was monitored on a strip chart recorder for approximately 30 min. Seawater containing algae (0.5 mg/ℓ) was pumped into the vessel during an acclimation period at a rate of 80 ml/min to ensure that food was present in the chamber and that routine metabolic rate was measured.

51. Absorption efficiency. After completion of the respiration rate measurements, all fecal material was removed from each feeding chamber. This ensured that only the algae consumed during the SFG procedures were used in the absorption efficiency measurements. At the food concentration used in the SFG measurements, approximately 0.5 mg/ℓ, no pseudofecal production occurred. The mussels were allowed to feed overnight in the chambers, and the feces were collected from each chamber the following morning. Fecal material was dried for 24 hr at 100° C, weighed, ashed at 500° C for 4 hr, and reweighed to determine the ash-free dry weight:dry weight ratio. A similar procedure was completed with the cultured algae to obtain the ash-free dry weight:dry weight ratio of the food. Absorption efficiencies were calculated for each treatment according to the method of Conover (1966).

52. Ammonia excretion rate. Mussels were placed individually into HCl-stripped beakers containing 300 ml of 1-μ filtered seawater for a period of 3 hr. Mussels were then removed and a 0.45-μ filtered, 50-ml sample was collected from each beaker, deposited into acid-stripped polyethylene bottles, and stored in a freezer at -20° C until analyzed. Ammonia analyses were completed in duplicate for each sample according to the method of Bower and Holm-Hansen (1980).

SFG calculations

53. After completion of the physiological measurements, the length and volume of each mussel were measured and the tissue excised, dried for 24 hr at 100° C, and weighed. The clearance rates, respiration rates, and ammonia

excretion rates were standardized to the mean weight of all the mussels used in the treatment. This procedure was used instead of standardizing to a 1-g mussel by using allometric equations because all mussels were approximately the same length and weight. The use of allometric equations is necessary only when mussels of variable size and weight are used (Bayne, Clark, and Moore 1981).

54. The weight-standardized values for each mussel were then used to calculate the SFG of each individual by substitution into the following equation:

$$SFG = (C \times A) - (R + E) \tag{5}$$

where

C = energy consumed (clearance rate × surrounding food concentration × energy of food)

A = absorption efficiency

R and E = energy lost through respiration and nitrogen excretion, respectively

The following energy conversions were used to calculate SFG:

1 mg of *T-Iso* = 4.5×10^7 cells (this experiment)
1 mg of *T-Iso* = 19.24 J (this experiment)
1 ml O_2 respired = 20.08 J (Crisp 1971)
1 mg NH_4-N = 24.56 J (Elliot and Davidson 1975)

The energy content of *T-Iso* was determined by filtering a volume of the algae onto preweighed glass fiber filters, drying them at 100° C for 24 hr, and reweighing them to determine algal dry weight. They were analyzed then using the dichromate wet oxidation method of Maciolek (1962) to determine oxygen consumed and the resultant energy content.

Clearance rates in
<u>laboratory exposure system</u>

55. In the laboratory documentation portion of the FVP (Nelson, Black, and Phelps 1985) it was noted that the main factor contributing to lowered SFG values in BRH-exposed mussels was decreased clearance rates. Therefore, it was decided to monitor the clearance rates of mussels in the exposure chambers during this set of experiments in order to observe when an adverse biological effect first began. The clearance rates were estimated using transmissometer readings (TRNUM) in each exposure chamber. As previously described, the TRNUM

was calibrated to the level of suspended particulates in each chamber. An initial transmissometer reading was taken for each exposure chamber. The dosing valve was then shut off for a period of 10 min, after which the final TRNUM was recorded. In this manner the decrease in suspended particulate level (milligrams/litre) was determined as the difference between the initial and final concentrations. This value was multiplied by the chamber volume (100 ℓ) to determine the total milligrams of material removed from the chamber. In addition to suspended particulates being removed by the mussels, some material was lost down the overflow. This was accounted for by multiplying the flow rate (0.4 ℓ/hr) by the mean particulate level during the 10-min period. This value was subtracted from the total volume removed to determine the volume of material removed by the mussels (MGS). Because of the design of the chamber, very little settling out of suspended sediment occurred, so this possible route of sediment loss during the 10-min time period was not factored in. The clearance rate (CR) was then calculated using the following formula:

$$\text{CR (litre/hour/mussel)} = \text{MGS} \times \frac{60\ \text{min/hr}}{10\ \text{min}} \div \begin{matrix}\text{Mean}\\ \text{mg}/\ell\end{matrix} \div \begin{matrix}\text{Total number}\\ \text{mussels}\\ \text{per chamber}\end{matrix} \qquad (6)$$

The clearance rates were measured in this manner for each treatment on Day 9 in the first experiment and Days 7 and 16 in the second experiment. The CR measurement on Day 16 was taken twice. The first one was completed immediately after the exposure tanks were cleaned when it was observed that the algae pump was not on. The pump was then turned on, the system allowed to equilibrate for 1 hr, and the CR measurement repeated. Both of these measurements were included to show the possible loss of an initial "active feeding" response in the mussels from the 30-percent BRH treatment when food was first provided.

Actual growth

56. The actual growth of 10 mussels was measured from each treatment for comparison with SFG values. This was accomplished by numbering 15 mussels (extras included in case of mortality) in each treatment prior to the start of the experiment, recording the length, and remeasuring the mussels on Day 14 in the first experiment and Days 14 and 28 in the second experiment. In this manner, growth data were obtained separately for the first and second 14-day periods in the second experiment.

Field Methods

Organism collection and holding

57. All mussels used in the field studies for the FVP were collected by scallop dredge from Narragansett Bay. In general, *M. edulis* were collected 1 to 2 days prior to field deployment to Long Island Sound. They were returned to the laboratory where 100 5- to 7-cm organisms were sorted and placed into polyethylene baskets. Each basket was placed in holding tanks of flowing unfiltered seawater until deployed in the field.

Exposure

58. *Mytilus edulis* were deployed at CNTR, 400E, 1000E, and REFS at the CLIS disposal site (Figure 2). The physical arrangement of each station is detailed by Phelps and Galloway (1980). In short, each station consisted of a surface buoy attached by cable to a concrete mooring on the bottom, with two smaller satellite moorings attached to the larger main mooring. A subsurface buoy was attached to each small mooring from which the mussel baskets were hung 1 m above the bottom. Two baskets were attached to each subsurface buoy at each deployment.

59. The deployments of *M. edulis* at the CLIS disposal site are summarized in Table 2. Mussels were deployed at each station for a period of 1 month predisposal to collect baseline data (Cruise number T - 04). A second deployment occurred during disposal operations, except that no mussels were placed at the CNTR station (T = 0, T + 2). Mussels were deployed for 1-month periods over the next 3 months (T + 8, T + 12, T + 15), then on a quarterly basis for the next year (T + 27, T + 43, T + 74, and T + 116). In addition, several sets of mussels were left at each station for 7 months (T + 21). The cruise number designation is not related to the length of deployment.

60. *Mytilus edulis* were retrieved from the subsurface buoys by divers. Mussels used for chemical analysis were frozen immediately. The remaining mussels were maintained in tanks of flowing seawater on deck and returned to ERLN later that day and held in flowing unfiltered seawater overnight. The next morning, mussels were distributed to the appropriate investigators for biological analyses.

61. Estimated field exposure via tissue residues. Exposure conditions present in the field during each mussel deployment were not as well characterized as they were in the laboratory studies. As a result, the description

Table 2

Cruise Number, Deployment Date, Retrieval Date, and Length of Deployment for Mussels Transplanted to CLIS

Cruise Number Weeks*	Deployment Date	Retrieval Date	Length of Deployment
T - 04	16 Mar 83	22 Apr 83	1 month
T = 0**	22 Apr 83	24 May 83	1 month
T + 2	23 Apr 83	07 Jun 83	6 weeks
T + 8	07 Jun 83	13 Jul 83	1 month
T + 12	13 Jul 83	10 Aug 83	1 month
T + 15	10 Aug 83	06 Sep 83	1 month
T + 21	16 Mar 83	18 Oct 83	7 months
T + 27	06 Sep 83	29 Nov 83	3 months
T + 43	29 Nov 83	20 Mar 84	4 months
T + 55	18 Oct 83	05 Jun 84	8 months
T + 74	12 Jun 84	17 Oct 84	4 months
T + 116	11 Jul 85	14 Aug 85	1 month

* The "cruise number, weeks" is not related to the "length of deployment."
** T = 0 refers to the termination of disposal activities at the FVP site on 18 May 1983.

of *M. edulis* exposure to BRH material in the field is more qualitative than quantitative and will be presented in two parts. First, a prediction of field exposure is based on mussel tissue residues. The relationship between exposure to BRH sediments and tissue residues was determined in the laboratory experiments. Tissue residues from the 0-, 10-, and 30-percent BRH treatments at 28 days were regressed against measured BRH exposure concentrations (0, 1.5, and 3.3 mg/ℓ) from the same exposures. In order to correct for background residues in the laboratory, the PCB concentration of the 0-percent BRH treatment was subtracted from the others prior to regression analysis. The resultant equation, milligrams per litre of BRH material = (PCB residue × 0.000965) - 0.0019 , $R^2 = 0.999$, was then used to calculate the average sustained concentration of BRH material necessary to achieve the residue value obtained in the field. The estimated BRH exposures in the field were determined by substituting the mussel PCB tissue residue concentration directly into the above equation. This estimate was assumed to represent an upper range of suspended BRH material present. A second estimate was determined by first subtracting the PCB concentration in mussels at the REFS station from the other stations during that collection. This removed the Long Island Sound background PCB levels from the estimates, and thus was assumed to represent a lower range of BRH present in CLIS. This procedure was completed for each collection date and station at which mussels were retrieved.

62. Estimated exposure via water chemistry data. A second estimate of exposure was generated from the PCB and Cu concentrations in the whole water samples collected during various postdisposal cruises. The concentration of BRH material that would have to be present to produce these levels was determined by dividing the concentration of PCB and copper present in the barrel material collected from BRH (2,900 μg/g and 6,910 ng/g for Cu and PCB, respectively). A range of exposures was also calculated for the water chemistry data; estimated BRH material was determined with and without subtracting the concentration at the REFS station.

SFG procedures

63. SFG was measured the morning after mussels were returned from the field. The SFG procedures used to analyze the field mussels were the same as those used to measure the laboratory mussels. Physiological measurements of the field-collected mussels were conducted at the same ambient temperature they were exposed to in the field. Ambient water temperatures in Narragansett Bay were within 1° to 2° C of those at the CLIS disposal site.

Statistical analysis

64. The primary objective of the FVP was to compare laboratory and field responses under similar conditions. The highly dynamic temporal and spatial conditions in the field made it impossible to replicate these conditions in the laboratory. Consequently, the experimental design employed was such that qualitative relationships were made between the laboratory and field. Therefore, descriptive statistical procedures were used, not inferential tests. Standard errors presented in tables and figures were calculated from 10 samples within a treatment or basket. These values are included to indicate variability within a treatment, not among 10 statistical replicates.

65. Statistical analysis of the data was completed in three parts, exposure-effects, residue-effects, and laboratory-to-field comparison. In the laboratory experiments, regression analysis was used to determine the

relationship between SFG and BRH exposure concentration (Snedecor and Cochran 1967). The limited exposure data from CLIS precluded measuring similar relationships for the field mussels. The relationship between SFG and tissue residue in the laboratory experiments was determined by regressing the mean SFG value for each treatment against the corresponding mean tissue residue (Snedecor and Cochran 1967). Data from both laboratory experiments were included. This procedure was completed individually for each of the 10 selected chemical contaminants and the two summary statistics.

66. Prior to making comparisons between laboratory and field effects, it was necessary to establish whether exposures (i.e., residues) were similar in the laboratory and the field. This was accomplished by examining the tissue residues of all mussels from laboratory and field exposures together, independent of exposure concentration or station location and date. The PCB, ethylan, PAH, and SUM and CENT variables were analyzed by cluster analysis (BioMedical Programs 1983) to establish which tissue residues, among all the laboratory treatments and field stations, were most similar. Prior to analysis, residue values for each compound were normalized using standard deviations from the mean. This procedure ensured that each variable was weighted equally, even if its absolute value was orders of magnitude different from another variable.

PART III: RESULTS

Laboratory

Exposure

67. System monitoring. The *M. edulis* exposure system was monitored for both total suspended solid (TSS) concentrations and the percentage of REF and BRH sediments. The strip chart record indicated that the system maintained a suspended particulate concentration of 10 mg/ℓ approximately 90 percent of the time. Examples of times when the 10 mg/ℓ was not maintained include periods when exposure tanks were cleaned, slurry reservoirs were changed, and lines were clogged. Overall, the system provided a nearly constant total suspended particulate concentration to the mussels. The concentration of BRH sediments dosed into each treatment is listed in Table 3.

Table 3

Suspended Sediment Concentrations in the Mussel Exposure System*

Nominal Percent BRH	Measured Percent BRH**	Calculated BRH Sediment mg/ℓ
100	100 (0.00)	10.0
50	50 (0.83)	5.0
30	33 (0.84)	3.3
10	15 (1.39)	1.5
0	0 (0.00)	0.0

* Values include nominal and actual BRH suspended sediment concentration.
** Standard error in parentheses.

68. When the TSS concentration dropped in the 50-percent BRH exposure tank, a pulse of equal length was sent to both the REF and BRH dosing valves. Volumetric measurements of the BRH and REF sediment doses indicated that equal amounts (±5 percent) of BRH and REF material were delivered to the 50-percent BRH exposure chamber.

69. The 10- and 30-percent BRH treatments required two dosing valves per treatment. Because the pulse length could not be adjusted separately for

each valve, manual adjustment of each valve was required to provide the desired percent concentration. The volumetric amount of BRH and REF material delivered to each treatment was monitored and recorded (Table 3). In the treatment with a nominal concentration of 10-percent BRH, the actual value delivered was 15 percent. In the 30-percent BRH treatment, the actual value was 33 percent BRH.

70. Chemical monitoring. The results of the chemical monitoring are prefaced by a brief restatement of the purpose of the exposure system to aid in the understanding of the results. The system used in this experiment was designed to maintain a constant particulate concentration of 10 mg/ℓ in the exposure chambers. Initially, 150 animals were placed into each chamber with clearance rates of approximately 2 ℓ/mussel/hr, or a total of 300 ℓ/hr. The seawater flow rate through each chamber, independent of suspended sediment additions, was approximately 24 ℓ/hr. In effect, suspended sediment was added at a rate 12.5 times that of seawater to each exposure chamber each hour to compensate for sediment removed by the mussels. This has important consequences on the behavior of the contaminants in the exposure system.

71. If all the contaminants were associated with the suspended sediment, the contaminant concentrations in the exposure chambers should be similar to those predicted by regressing the TSS concentrations with contaminant concentrations in the BRH material. Conversely, any contaminants that do not remain bound to the particulates could attain concentrations in the exposure system different from those predicted from the TSS data. This occurs because the mussels in the system are more efficient at removing the particulate-bound contaminants than they are at removing the dissolved contaminants. This theory is proposed to explain the results of the chemical concentrations in the exposure system, using PCB and copper as examples.

72. Whole water samples were taken for chemical analysis on Days 1, 7, 14, 21, and 28 in the second experiment. The mean PCB concentrations (nanogram/litre) for the five sampling dates for each exposure treatment in the second experiment are given in Table 4. The corresponding concentration of BRH sediment was estimated by regressing the nominal concentration of BRH against the expected value of PCB. Expected PCB concentrations were based on the PCB concentrations in the BRH sediment (6 ng/mg) plus background seawater concentrations. Substitution of the actual measured values of PCBs in the exposure system into the equation provided an estimated value of the

Table 4

Chemical Monitoring of the Exposure System in Experiment 2

Nominal Treatment Concentration, % BRH	PCB Concentration, ng/ℓ		BRH Concentration, mg/ℓ	
	Expected	Measured	Estimated	Measured
0	1.1	2.2	0.2	0
10	7.1	11.9	1.8	1.5
30	18.8	23.6	3.8	3.3

concentration of BRH sediment in the system. The estimated concentration of BRH sediment in each treatment is similar to the actual measured values. These data suggest that PCB concentrations in the system are closely associated with the TSS concentrations.

73. Copper concentrations were measured both with and without mussels in the exposure system at 10 mg/ℓ TSS for each treatment. With no mussels in the exposure system, the total copper concentrations were 9.37 and 2.5 μg/ℓ for the 30- and 10-percent BRH treatments, respectively. These concentrations represent 3.8 and 1.8 mg/ℓ BRH sediment in the two treatments, respectively. Under these conditions, the predicted and measured copper concentrations were comparable. This resulted because the effective flow of suspended sediment and incoming seawater is the same. The only loss of TSS was out the overflow due to seawater flow rates.

74. When mussels were present in the system, the mean copper concentrations were 17.0 and 10.7 μg/ℓ for the 30- and 10-percent BRH treatments, respectively. These copper concentrations correspond to 68- and 43-percent BRH sediment in the two treatments, respectively, and conflict with those expected from the TSS data. The results may be explained by the fact that copper, due to its solubility in seawater, became disassociated from the TSS because suspended solids were delivered at a higher rate to the exposure chamber than the rate of incoming seawater. When a dose of BRH suspended sediment was delivered to an exposure chamber, all contaminants were introduced at the same rate. Because the mussels were more efficient at removing particulates than dissolved contaminants, dissolved copper tended to accumulate, which resulted in higher concentrations of copper than those predicted from the TSS data alone.

Tissue residue

75. Differences in contaminant concentrations between BRH and REF sediments facilitated the tracking of these contaminants in exposed biota (Appendix B). Results of Experiment 1 indicate that PCB tissue concentration in mussels is directly related to exposure concentration (Table 5). PCBs in mussels from the 0-percent BRH concentration remained about the same over the 14-day experiment.

Table 5

PCB Tissue Residues (ng/g dry weight) in Mussels from Laboratory Experiment 1

Day	PCB Residue in Indicated Percent BRH Sediment		
	0	50	100
0	117	117	117
14	154	2,100	3,700

76. The PCB residue data from Experiment 2 are listed in Table 6 and graphically depicted in Figure 6. Tissue residues, measured at 7-day intervals, indicated that the mussels in the 0-percent BRH chamber maintained a relatively constant background concentration of PCBs throughout the experiment. In the 10- and 30-percent BRH chambers, the concentration of PCBs in the mussels increased from Day 0 to Day 14, then remained nearly constant

Table 6

PCB Tissue Residues (ng/g dry weight) in Mussels from Laboratory Experiment 2

Day	PCB Residue in Indicated Percent BRH Sediment		
	0	10	30
0	210	210	210
7	280	1,110	2,100
14	270	1,910	3,600
21	360	1,720	3,600
28	280	1,840	3,700

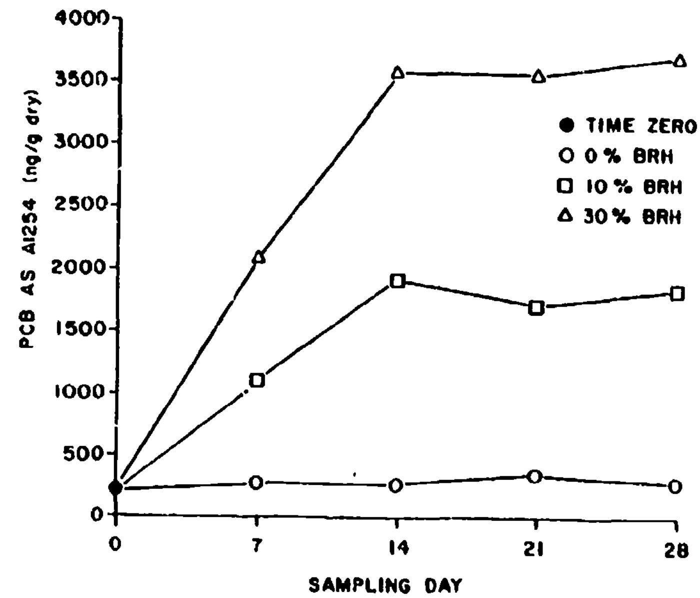


Figure 6. Concentrations of PCB as A1254 in the tissue of *M. edulis* exposed to BRH suspended sediment for 28 days

between Days 14 and 28, suggesting that the mussels reached a steady state somewhere between Days 7 and 14. The steady-state PCB concentration in the 30-percent BRH mussels was almost double that of the mussels from the 10-percent BRH treatment. The actual concentration of BRH dosed to the 30-percent BRH treatment, 3.3 mg/ℓ, is nearly double that dosed to the 10-percent BRH mussels, 1.5 mg/ℓ. The measured whole water PCB concentrations were 11.86 and 23.57 ng/ℓ for the 10- and 30-percent BRH treatments, respectively. These data indicate a good relationship between the actual dosed concentrations of BRH suspended sediment, the measured whole water concentrations, and the PCB tissue residues in the mussels in Experiment 2.

77. A comparison of the tissue residues between the two experiments can be made for Days 0 and 14. The PCB concentration in the Day 0 mussels from Experiment 1 was almost half that in Day 0 mussels from Experiment 2 (117 and 210 ng/g, respectively). In addition, Day 14 PCB concentrations were about the same for the 10- and 50-percent BRH exposed mussels (1,910 and 2,100 ng/g) as well as the 30- and 100-percent BRH exposed mussels (3,600 and 3,700 ng/g). These data show dose responses within each experiment; however, there is poor agreement between experiments. The PCB data from these experiments were normalized to nanograms/gram of lipid, and the results are presented in Table 7

Table 7

PCB Concentrations (ng/g Lipid) in Mussels from Both Laboratory Experiments

Day	PCB Concentration in Indicated Percent BRH Sediment					
	0*	0**	10**	30**	50*	100*
0	2,900	2,400	2,400	2,400	2,900	2,900
7	--†	5,200	17,100	24,000	--	--
14	3,800	4,300	27,000	54,000	53,000	119,000
21	--	5,000	35,000	67,000	--	--
28	--	3,800	30,000	66,000	--	--

* Experiment 1.
** Experiment 2.
† Not sampled.

and Figure 7. Inspection of these data shows that differences between experiments can be explained when differences in lipid content of the organisms are taken into account. In addition, this procedure indicates that a dose-response relationship does exist between experiments when the Day 14 data from both experiments are combined (Figure 7).

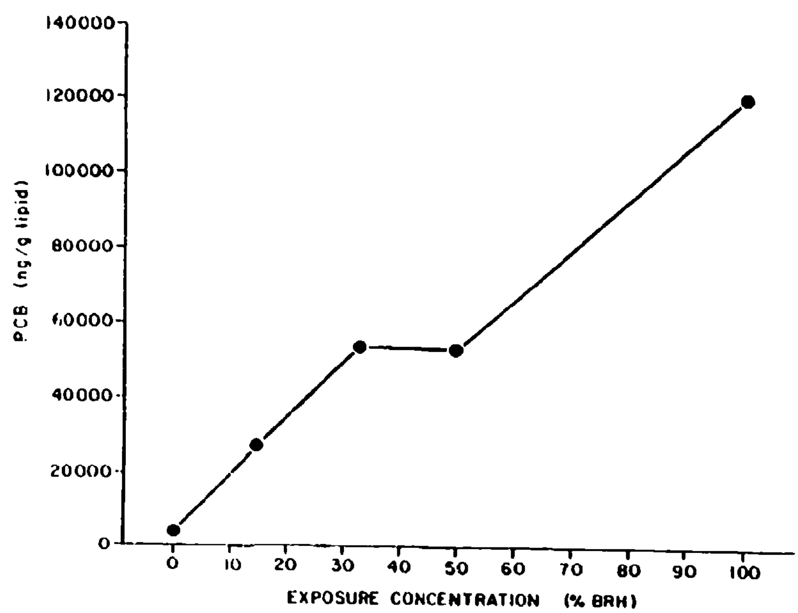


Figure 7. Concentrations of PCB as A1254, normalized for lipids, in the tissue of *M. edulis* exposed to BRH sediment for 14 days

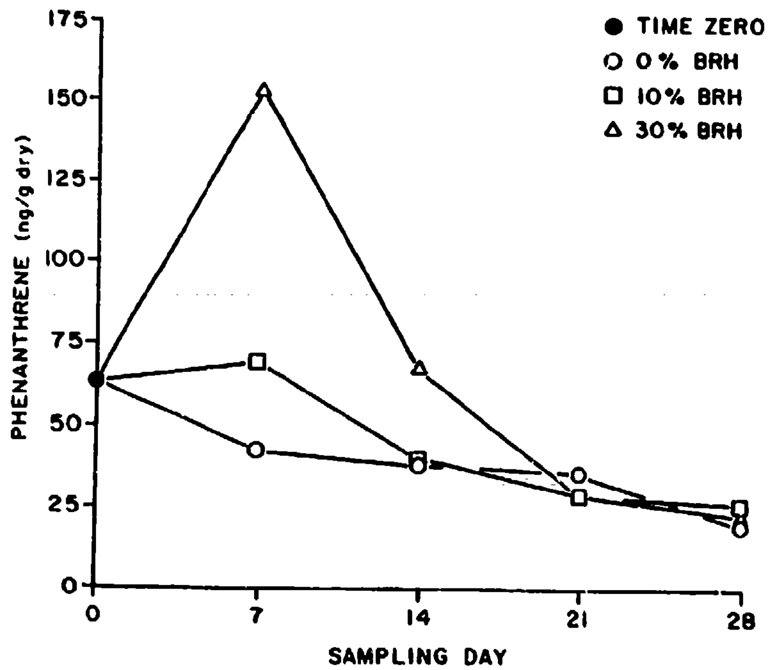
78. In addition to PCBs, tissue residues of phenanthrene, the sum of the 178 alkyl homologs, fluoranthene, benzo(a)pyrene, ethylan, cadmium, copper, chromium, and iron were also measured on Days 0, 7, 14, 21, and 28 of Experiment 2. The summary statistics, SUM and CENT, of the PAHs were also calculated for each of these sampling dates. These data are summarized graphically in Figures 8-13.

79. While each of the graphs presented as Figures 8-13 will not be discussed at length, it is interesting to note the relationship between the molecular weight of the organic compounds and tissue residue over time. The benzo(a)pyrene tissue residues follow a pattern similar to that of PCB. After 7 days, residues remain nearly constant for each exposure concentration. The fluoranthene residues are initially higher in the 30-percent BRH treatment. However, they decrease over the 28-day exposure period to a level comparable to the 10-percent BRH treatment. Residues for both of these treatments are elevated compared with the 0-percent BRH mussels. Phenanthrene, an even lower molecular weight PAH, increased initially but then decreased in both the 30- and 10-percent BRH treatments to a level comparable to the 0-percent BRH exposure. These data would suggest that mussels have the ability to metabolize and/or excrete the lower molecular weight PAHs, even during continuous exposure. In addition, the data would indicate that only higher molecular weight compounds should be used to relate exposure levels and subsequent tissue residue levels because, even under relatively constant exposure conditions, residues of lower molecular weight PAHs did not reflect exposure concentrations.

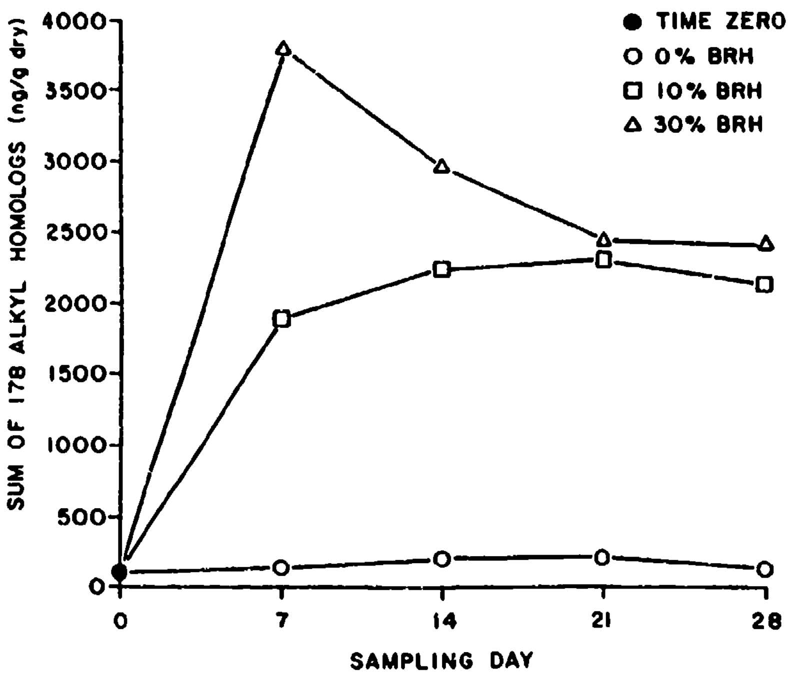
Biological effects

80. Clearance rates in the exposure system. Clearance rate measurements in the exposure system are listed in Table 8. In the first experiment, clearance rates were dramatically reduced in both the 50- and 100-percent BRH treatments after 9 days of exposure. This observation was reinforced by the strip chart record which indicated a reduced number of doses provided to these two treatments.

81. Similar results were observed in Experiment 2. Measurements on Day 7 indicated reduced clearance rates in both the 30- and 10-percent BRH treatments when compared with the 0-percent BRH treatment. On Day 16 this measurement was repeated soon after the exposure system was cleaned. The particulate levels were at the proper concentration; however, it was later noted

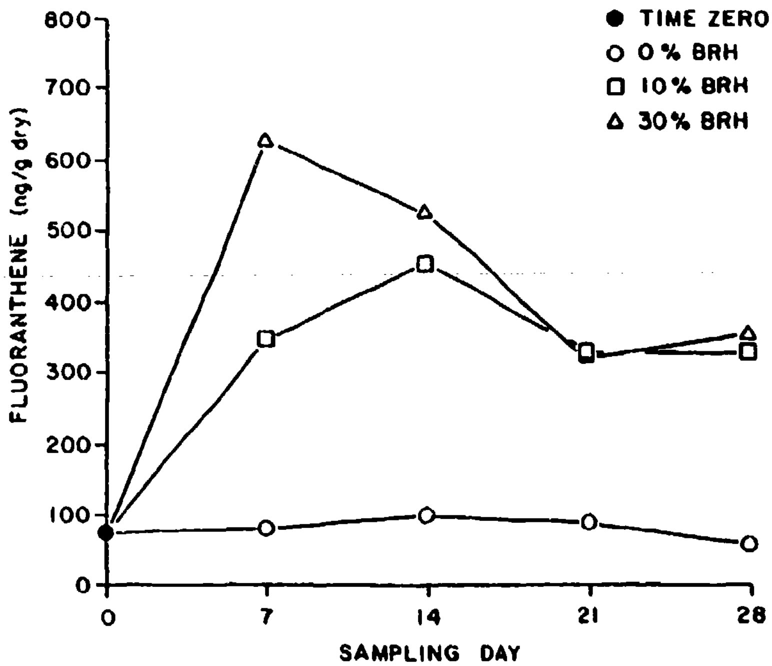


a. Phenanthrene

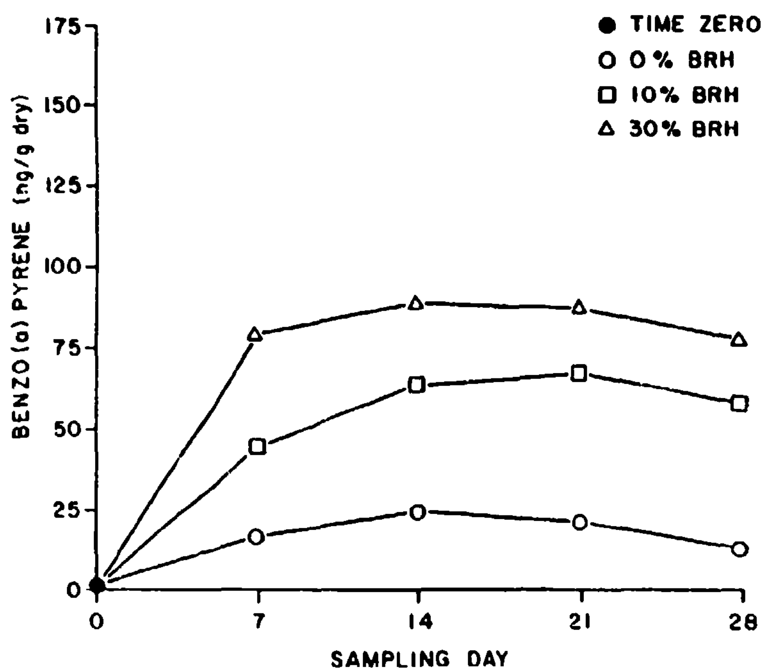


b. 178 alkyl homologs

Figure 8. Concentrations of phenanthrene and 178 alkyl homologs in the tissue of *M. edulis* exposed to BRH suspended sediment for 28 days

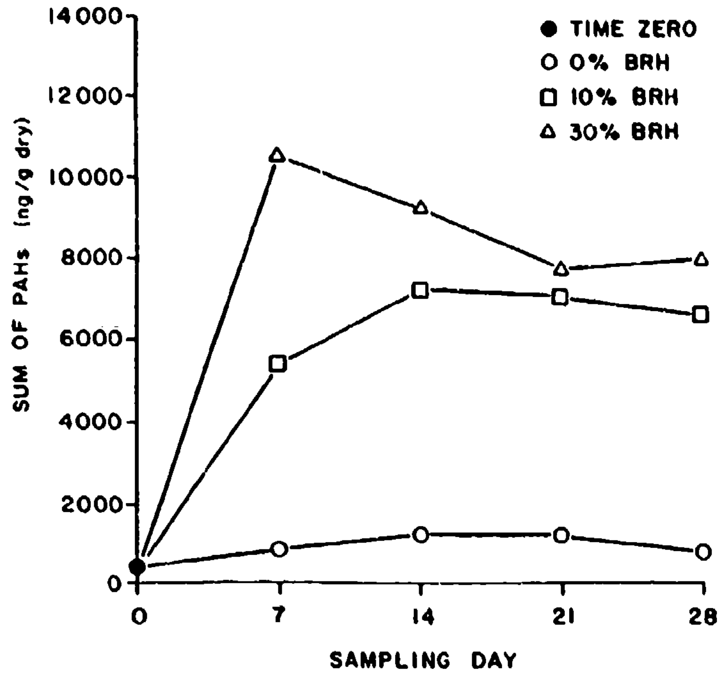


a. Fluoranthene

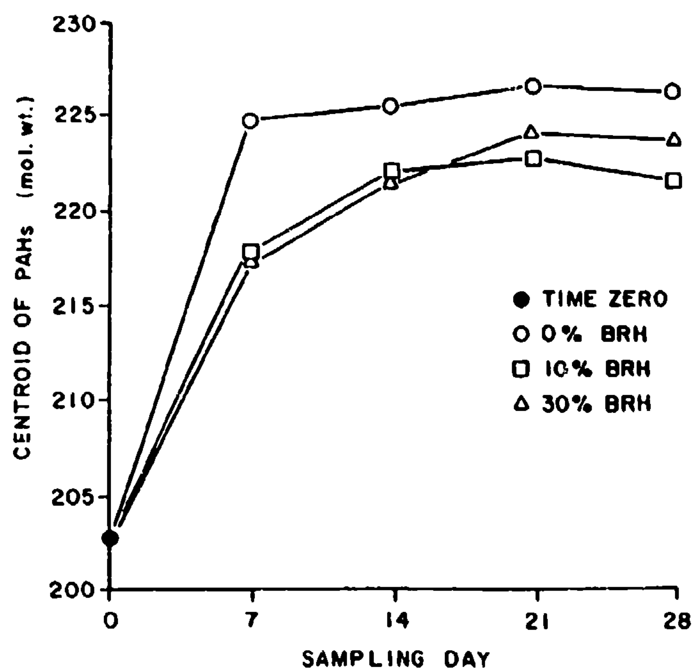


b. Benzo(a)pyrene

Figure 9. Concentrations of fluoranthene and benzo(a)pyrene in the tissue of *M. edulis* exposed to BRH suspended sediment for 28 days

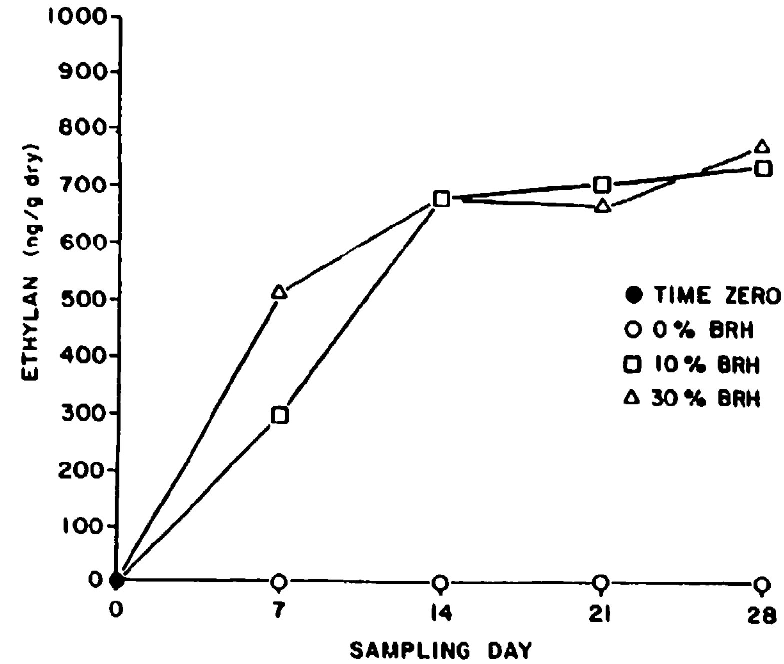


a. SUM of PAHs

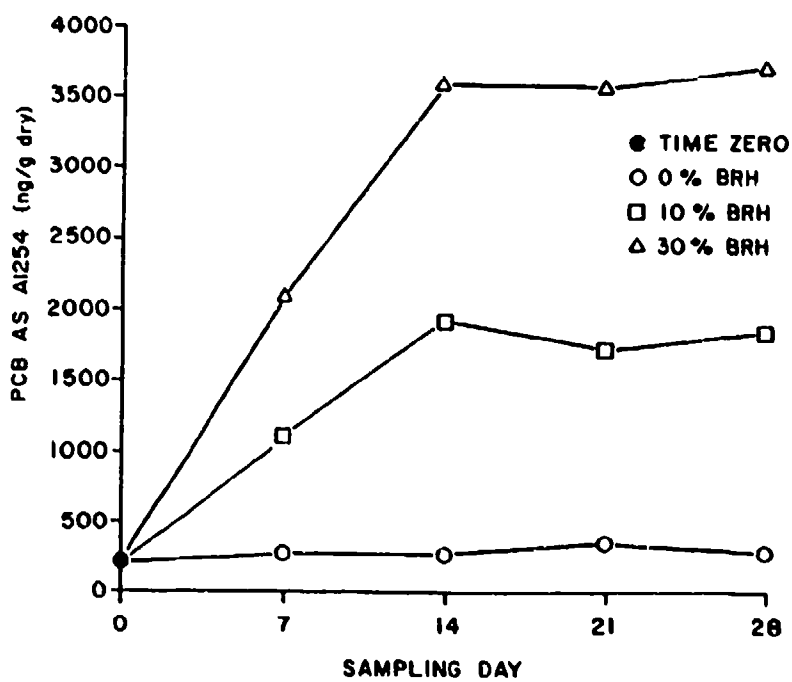


b. CENT of PAHs

Figure 10. Concentrations of the SUM of PAHs and CENT OF PAHs in the tissue of *M. edulis* exposed to BRH suspended sediment for 28 days

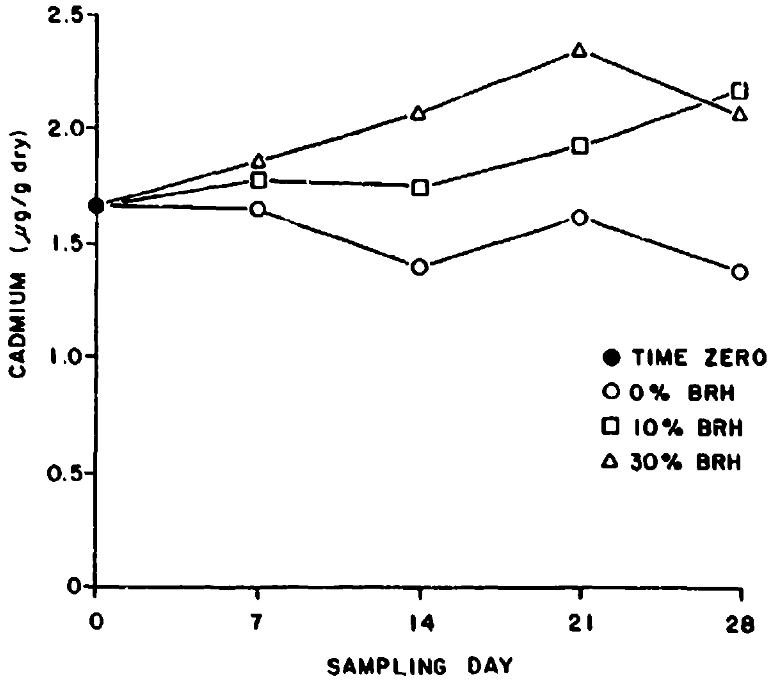


a. Ethylan

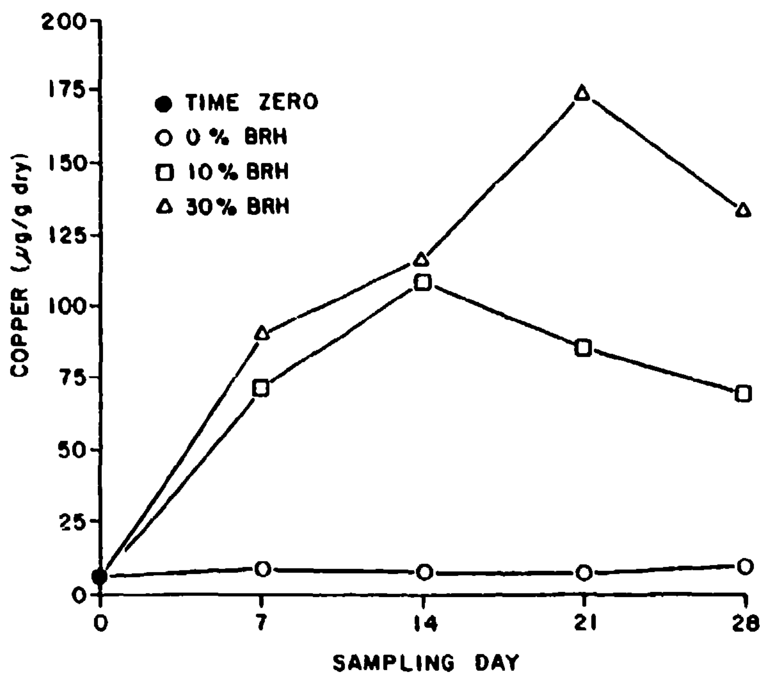


b. PCB

Figure 11. Concentrations of ethylan and PCB as A1254 in the tissue of *M. edulis* exposed to BRH suspended sediment for 28 days

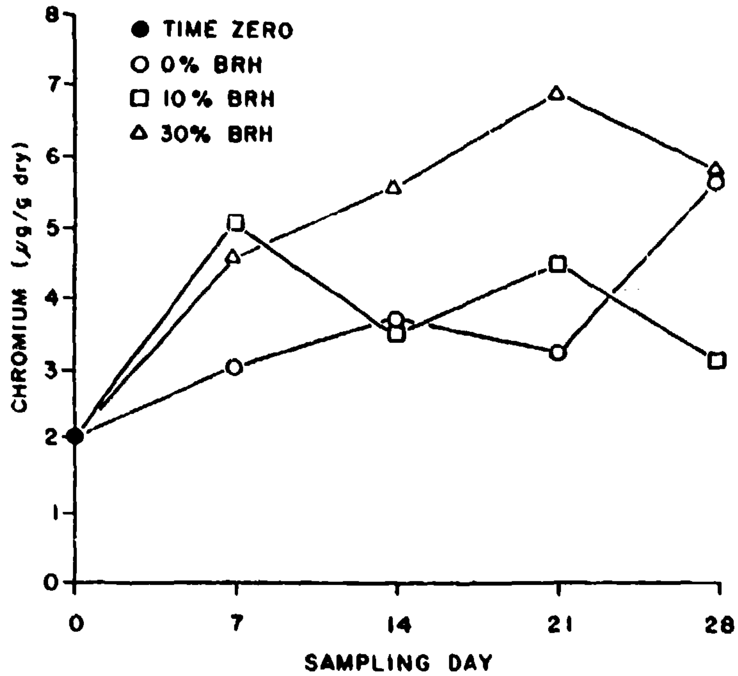


a. Cadmium

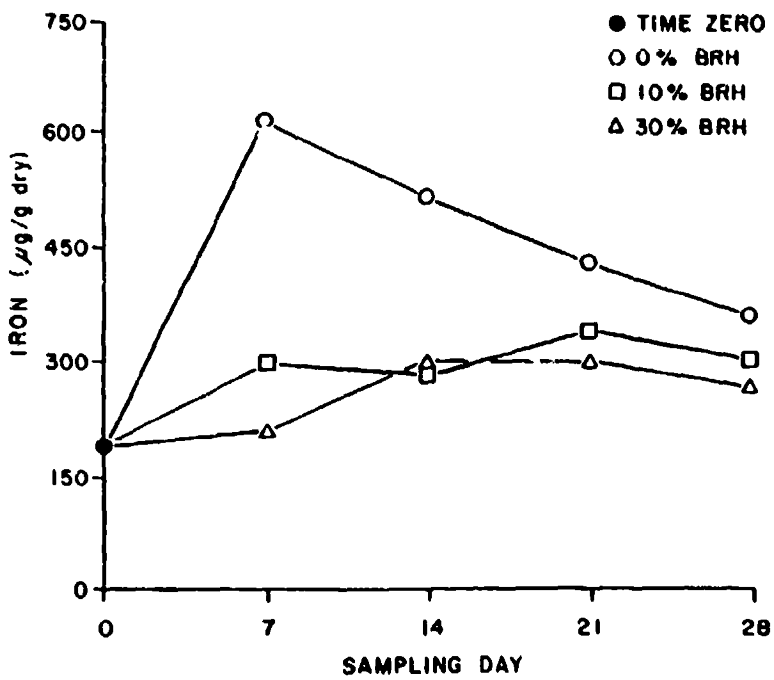


b. Copper

Figure 12. Concentrations of cadmium and copper in the tissue of *M. edulis* exposed to BRH suspended sediment for 28 days



a. Chromium



b. Iron

Figure 13. Concentrations of chromium and iron in the tissue of *M. edulis* exposed to BRH suspended sediment for 28 days

Table 8

Clearance Rates (litres/hr/mussel) of Mussels in Dosing System Exposure Tanks

Day	0% BRH	50% BRH	100% BRH
	Experiment 1		
9	2.22	0.10	0.10
	0% BRH	10% BRH	30% BRH
	Experiment 2		
7	1.60	0.52	0.26
16 (no algae)	1.23	0.90	0.82
16 (algae on)	2.87	2.55	0.94

that the pump supplying algae to the exposure tanks was off. At this time, the clearance rates in the 10- and 30-percent BRH treatments were again lower than the 0-percent BRH treatment. The algae pump was then turned on, and algae levels were allowed to equilibrate (about 1 hr). Clearance rate measurements were repeated, and this time the 0- and 10-percent BRH mussels both increased their clearance rates considerably to 2.87 and 2.55 ℓ/hr, respectively, while the clearance rate of the mussels in the 30-percent BRH treatment remained about the same as when no algae were present. Mussels are known to increase their feeding rate in the presence of food after being starved. Mussels from the 10-percent BRH treatment were able to increase their feeding rate to a level comparable to mussels from the 0-percent BRH treatment after the algae were added, while those in the 30-percent BRH chamber were not. These data indicate that mussels from the 30-percent BRH treatment were more severely impacted than those mussels from the 10-percent treatment.

82. SFG measurements. The values for the physiological parameters have been standardized to the mean weight of all the mussels for a particular experiment. The mean weight for the mussels from Experiment 1 was 0.48 g and for Experiment 2 was 0.74 g.

83. Clearance rates. The clearance rate data are listed in Table 9. The mussels from the 50- and 100-percent BRH chambers exhibited lower

Table 9

Clearance Rates of Mussels from the Two Laboratory Exposures*

Treatment % BRH		Clearance Rate ℓ/hr
	Experiment 1, Day 14	
0		4.69 (0.25)
50		0.54 (0.20)
100		0.17 (0.07)
	Experiment 2, Day 14	
0		4.47 (0.18)
10		2.48 (0.56)
30		0.81 (0.30)
	Experiment 2, Day 28	
0		3.51 (0.43)
10		1.80 (0.41)
30		1.07 (0.24)

* Each value represents the mean of 10 mussels (standard error in parentheses).

clearance rates after 14 days than the 0-percent BRH mussels in Experiment 1. In Experiment 2, on Day 14, mussels from the 30-percent chamber exhibited reduced clearance rates compared with the 10-percent BRH mussels, which were, in turn, lower than the 0-percent mussels. By Day 28, however, the clearance rates of mussels from the 30- and 10-percent BRH chambers were not much different from each other but were still lower than the 0-percent BRH mussels.

84. Absorption efficiency. Mussel absorption efficiencies are listed in Table 10. Only one pooled value was determined for each treatment. The results indicate that there were no large differences in absorption efficiency among the various exposure chambers.

85. Respiration rates. The respiration rate data are listed in Table 11. Differences among chambers were relatively small at all of the sampling times.

86. Ammonia excretion rates. The ammonia excretion rate values are listed in Table 12. The largest variations among the chambers occurred in Experiment 1, where the range of BRH exposure was the greatest.

Table 10

Absorption Efficiencies of Mussels from the Two Laboratory Exposures

Treatment % BRH	Absorption Efficiency, %
Experiment 1, Day 14	
0	78
50	80
100	85
Experiment 2, Day 14	
0	88
10	84
30	87
Experiment 2, Day 28	
0	72
10	78
30	85

Table 11

Respiration Rates of Mussels from the Two Laboratory Exposures*

Treatment % BRH	Respiration Rate ml O_2/hr
Experiment 1, Day 14	
0	0.41 (0.07)
50	0.39 (0.03)
100	0.41 (0.06)
Experiment 2, Day 14	
0	0.36 (0.02)
10	0.41 (0.03)
30	0.42 (0.02)
Experiment 2, Day 28	
0	0.38 (0.02)
10	0.48 (0.06)
30	0.46 (0.03)

* Each value represents the mean of 10 mussels (standard error in parentheses).

Table 12

Ammonia Excretion Rates of Mussels from the Two Laboratory Exposures*

Treatment % BRH	Excretion Rate µg NH_4-N/hr
Experiment 1, Day 14	
0	10.31 (0.90)
50	17.45 (3.09)
100	20.56 (10.48)
Experiment 2, Day 14	
0	12.69 (0.82)
10	11.48 (1.60)
30	11.33 (1.61)
Experiment 2, Day 28	
0	9.52 (1.37)
10	15.06 (2.34)
30	11.90 (1.39)

* Each value represents the mean of 10 mussels (standard error in parentheses).

87. SFG index. The Day 14 SFG values in the first experiment were reduced in the 50- and 100-percent BRH chambers (Table 13). The SFG values in the second experiment followed the same pattern as the clearance rates. By Day 14, mussels from the 10-percent BRH chambers exhibited lower SFG values than the 0-percent BRH mussels; however, their SFG values were higher than those of the mussels from the 30-percent BRH chamber.

88. The graph of the Day 14 SFG values and exposure concentration (Figure 14) suggested that the relationship between these two variables was not linear. Therefore, the SFG data were log 10 transformed prior to regression analysis. To avoid negative values (i.e., -7.14 for the 100-percent BRH treatment), each SFG number was increased by 8 prior to log 10 transformation. Regression analysis of the data indicated a significant inverse relationship ($P < 0.001$, $R^2 = 0.99$) between log SFG and BRH exposure concentration.

89. On Day 28, the SFG of mussels from the 10- and 30-percent BRH chambers were lower compared with those of the 0-percent mussels. However, they were not different from each other. Because there were only three data

Table 13

SFG Values of Mussels from the Two Laboratory Exposures*

Treatment % BRH	SFG (J/hr)
Experiment 1, Day 14	
0	10.62 (1.10)
50	-4.26 (1.54)
100	-7.14 (1.30)
Experiment 2, Day 14	
0	14.17 (0.59)
10	5.03 (2.00)
30	-2.82 (1.81)
Experiment 2, Day 28	
0	7.16 (1.86)
10	0.14 (1.30)
30	-1.79 (1.39)

* Each value represents the mean of 10 mussels (standard error in parentheses).

points, regression analysis was not appropriate. Visual inspection of the data suggested a relationship between SFG and BRH concentration similar to that observed on Day 14 (Figure 14).

90. Actual growth. In addition to the SFG determination, changes in shell length were measured on mussels from the same chamber (Table 14). In Experiment 1 the initial mean length of the mussels used was 5.02 ± 0.01 cm. The initial mean mussel length in Experiment 2 was 5.00 ± 0.01 cm. The growth increment is listed for the first 14-day period, in both experiments, and for the second 14-day period in the second experiment. In Experiment 1, shell growth of mussels from the 0-percent BRH chamber was greater than that of the mussels from the 50- and 100-percent BRH chambers. In Experiment 2, mussels in the 0-percent BRH chamber again showed a greater increase in shell growth than mussels from the 10- and 30-percent BRH treatments. Actual shell growth followed the same pattern as the SFG values and clearance rate measurements at Day 14 in both Experiments 1 and 2 and at Day 28 in Experiment 2 (Figures 15-17).

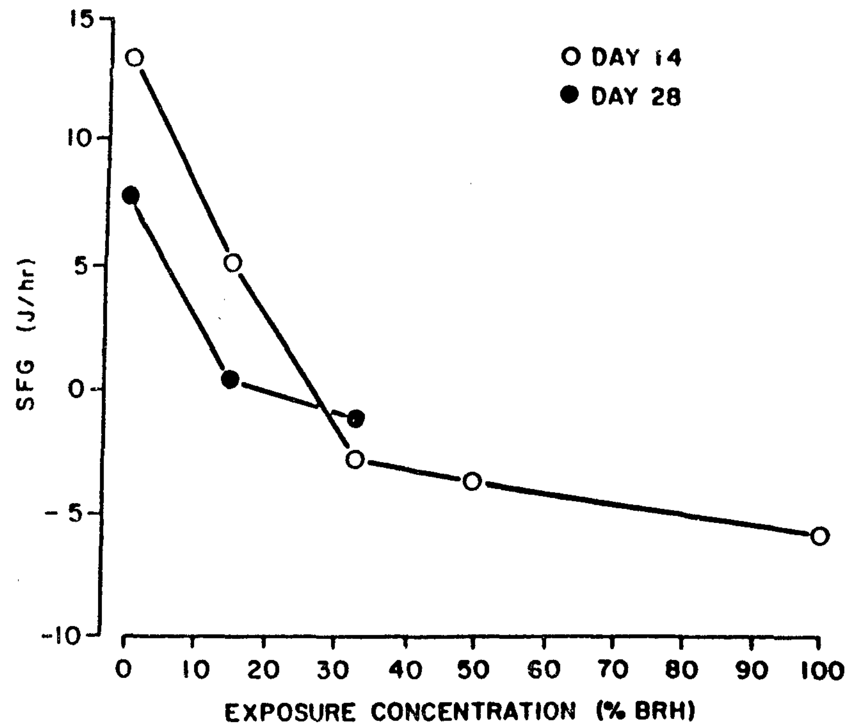


Figure 14. Relationship between the SFG of *M. edulis* and BRH exposure concentration on Days 14 and 28 of the laboratory experiments. Data from Experiments 1 and 2 are presented for Day 14

Table 14

Actual Growth of Mussels in the Exposure Chambers*

Treatment % BRH	Growth, mm Days 0-14	Growth, mm Days 15-28
	Experiment 1	
0	0.40 (0.13)	--
50	0.11 (0.06)	--
100	0.06 (0.04)	--
	Experiment 2	
0	0.75 (0.14)	0.73 (0.17)
10	0.41 (0.12)	0.28 (0.13)
30	0.07 (0.02)	0.04 (0.03)

* Values are the means of 10 mussels from each treatment (standard error in parentheses).

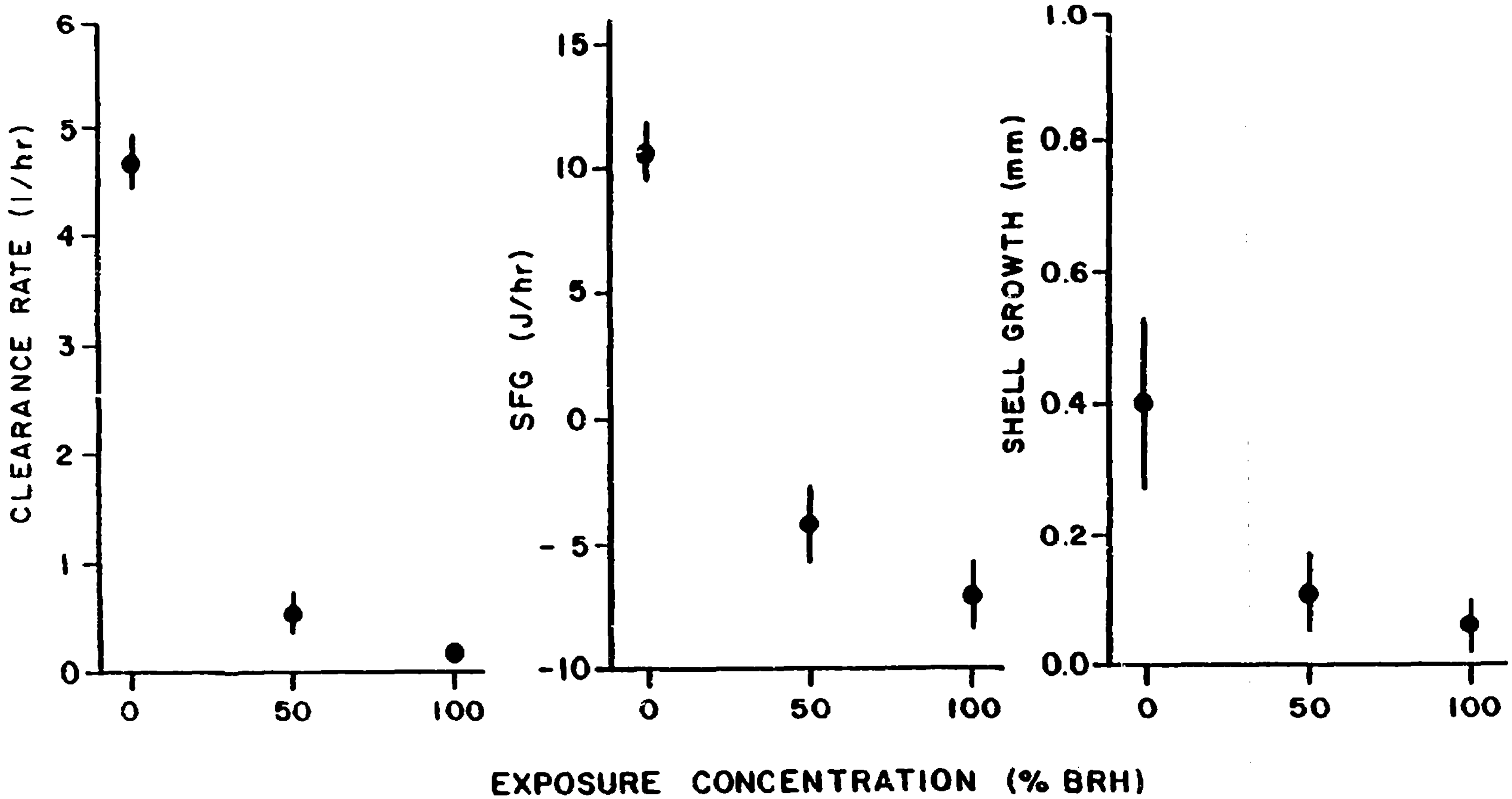


Figure 15. Effect of BRH suspended sediment on the clearance rate, SFG, and shell growth of *M. edulis*, Day 14, Experiment 1

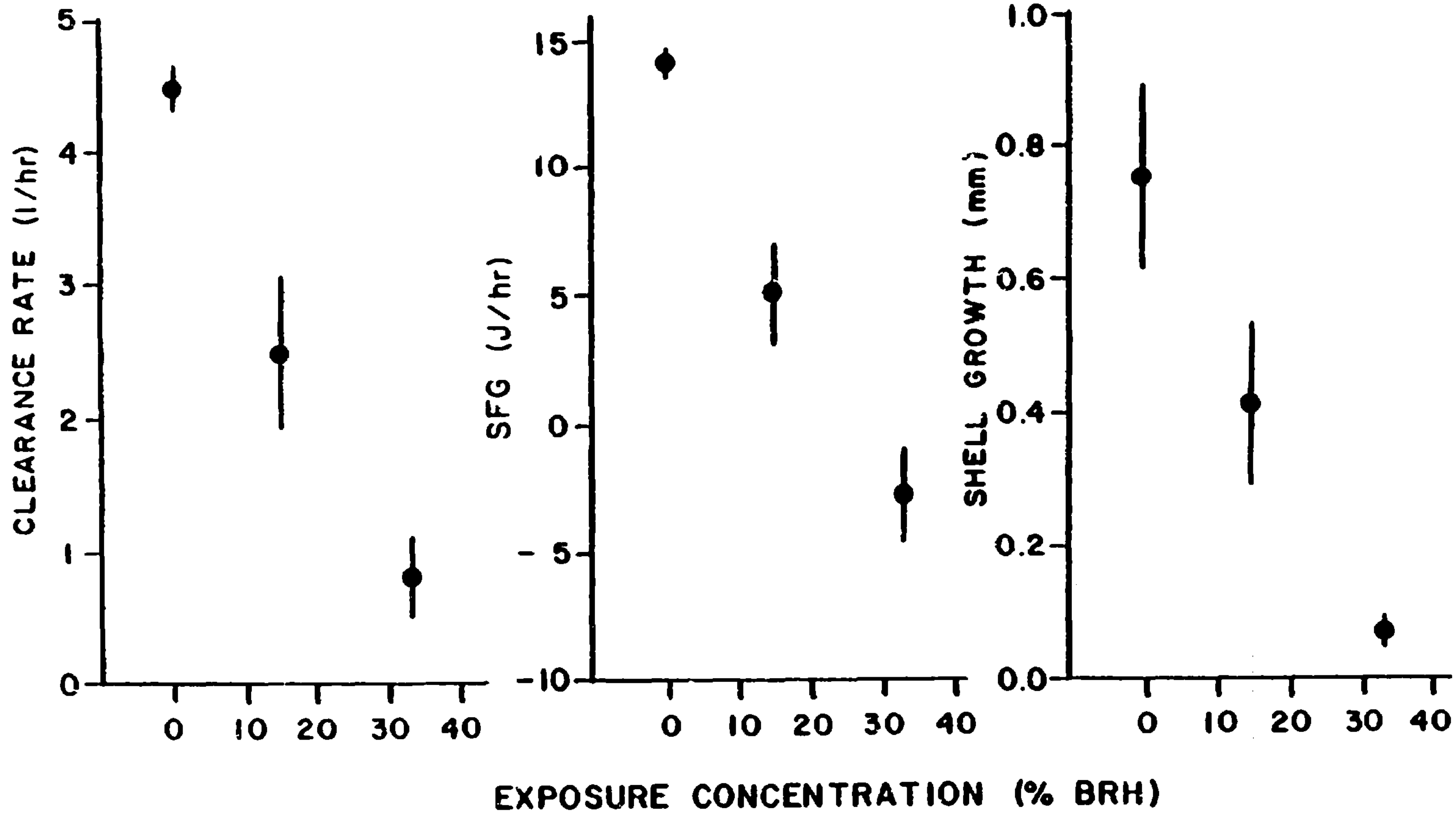


Figure 16. Effect of BRH suspended sediment on the clearance rate, SFG, and shell growth of *M. edulis*, Day 14, Experiment 2

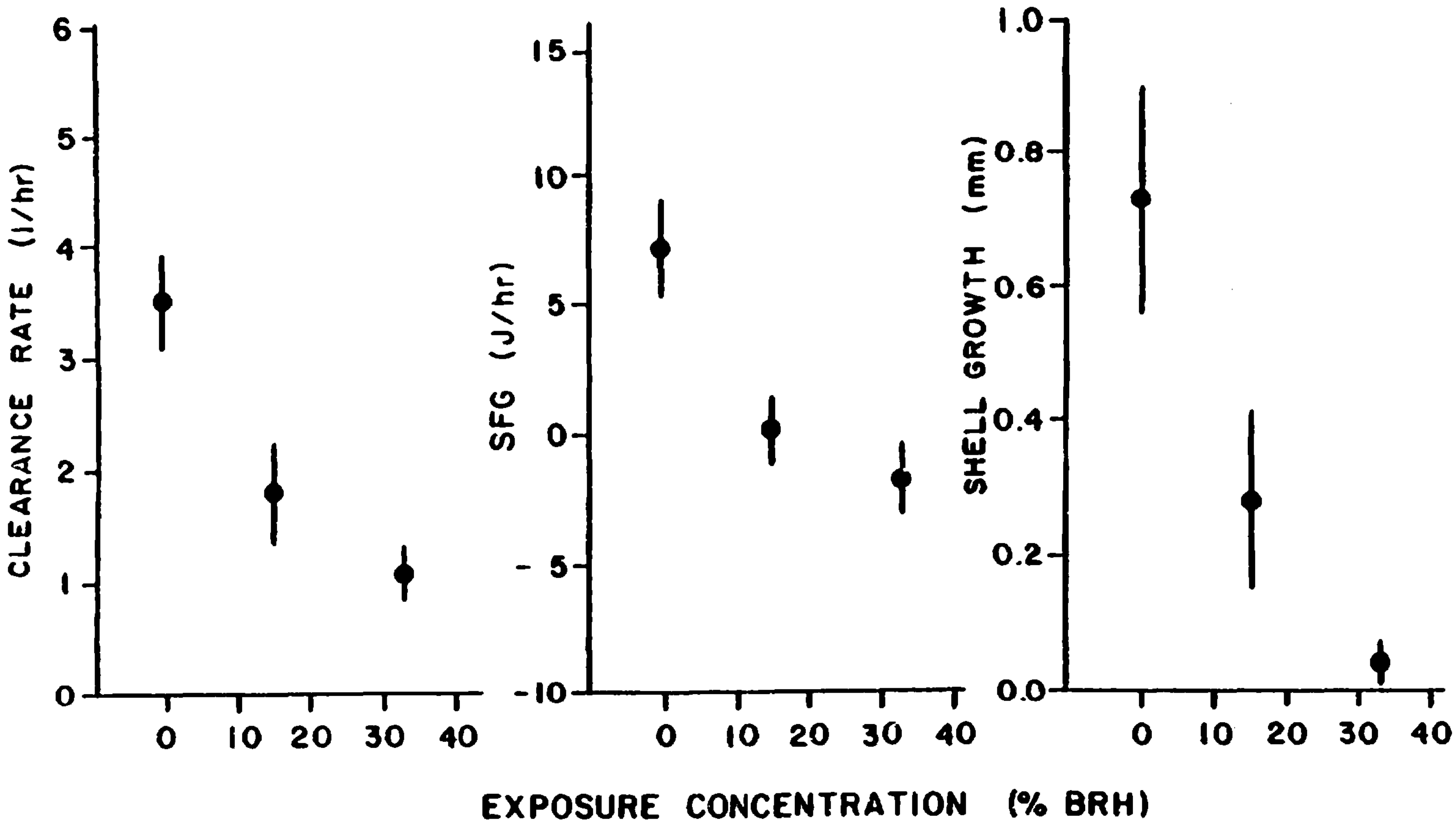


Figure 17. Effect of BRH suspended sediment on the clearance rate, SFG, and shell growth of *M. edulis*, Day 28, Experiment 2

91. In Experiment 2 it is also interesting to note the differences between the first and second 14-day growth periods. The mussels in the 0-percent BRH chamber grew virtually the same amount in the first 14 days as the second 14. In the 10-percent BRH chamber, growth was reduced in the second period as compared to the first. This was reflected in the SFG values as well (Table 13). In the 30-percent BRH treatment, very little growth occurred in either 14-day period.

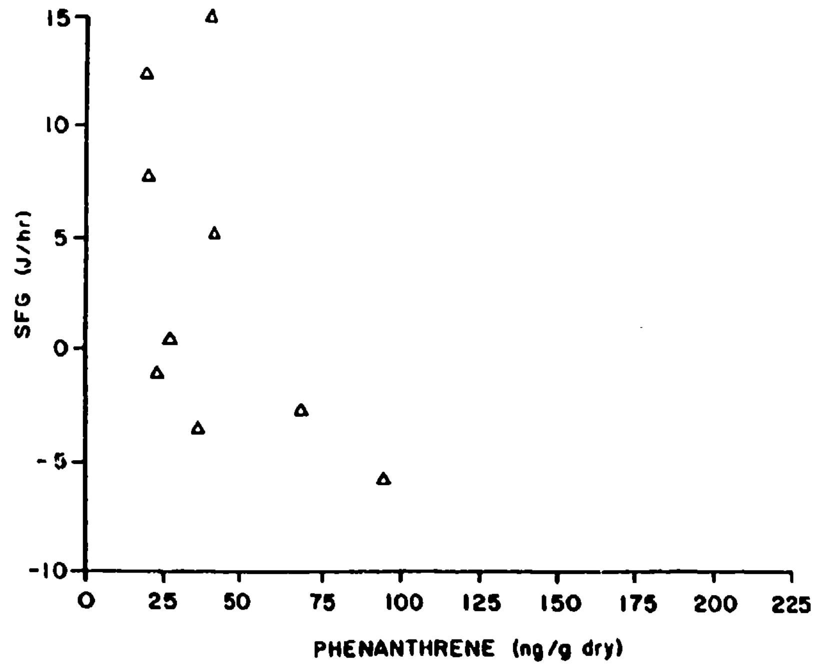
Residue effects

92. Regression analysis was used to determine whether any relationship existed between the SFG values and tissue residues. Data from both laboratory experiments were included because exposure conditions (i.e., temperature and total suspended particulate levels) were similar in each experiment; only the percent BRH differed between experiments and treatments. The relationship between SFG and tissue residue for each of the selected 10 chemicals, and two summary statistics, are presented graphically in Figures 18-23. The presence of a regression line on any graph indicates a significant relationship ($P < 0.05$) between SFG and the tissue residue for that particular element, compound, or summary statistic.

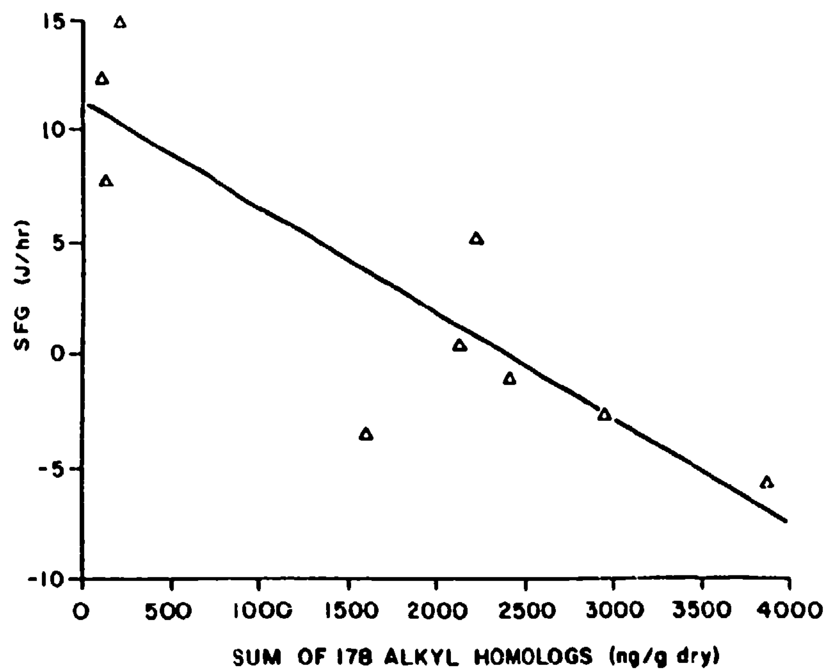
93. The individual graphs demonstrate that when a significant relationship exists between SFG and the tissue concentration of a chemical variable, that relationship is inverse in nature. These findings are consistent with the results of the residue-effects data presented above and support the contention that mussel residues accurately reflect BRH exposure. From the laboratory data for exposure, residue, and SFG, it is clear that increased exposure to BRH material is associated with increased tissue residue concentrations, reduced SFG, reduced clearance rates, and reduced growth in mussels.

94. Several chemical variables did not relate significantly to SFG values. Iron residues, for example, would not be expected to relate to SFG because iron concentrations are indicative of exposure to any sediment. Total suspended sediment levels were similar in all exposure concentrations.

95. The variable CENT showed no visible pattern to this distribution in mussels after exposure to different concentrations of BRH material, indicating that mussels may have accumulated only those contaminants which have a narrow range of octanol water partitioning coefficients (Lake, Hoffman, and Schimmel 1985). The other summary statistic, SUM, did show a significant negative relationship with respect to SFG. These data suggest that while the mussels

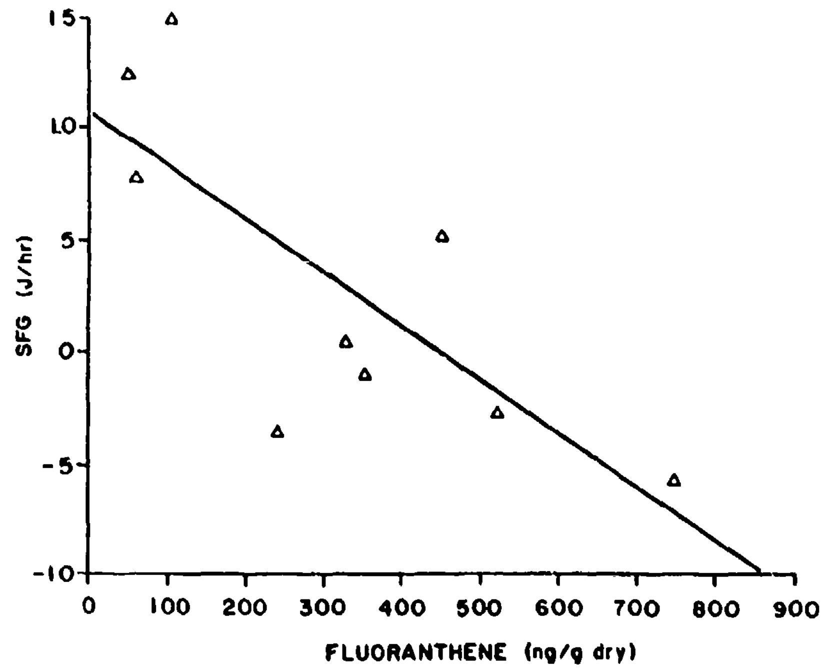


a. Phenanthrene

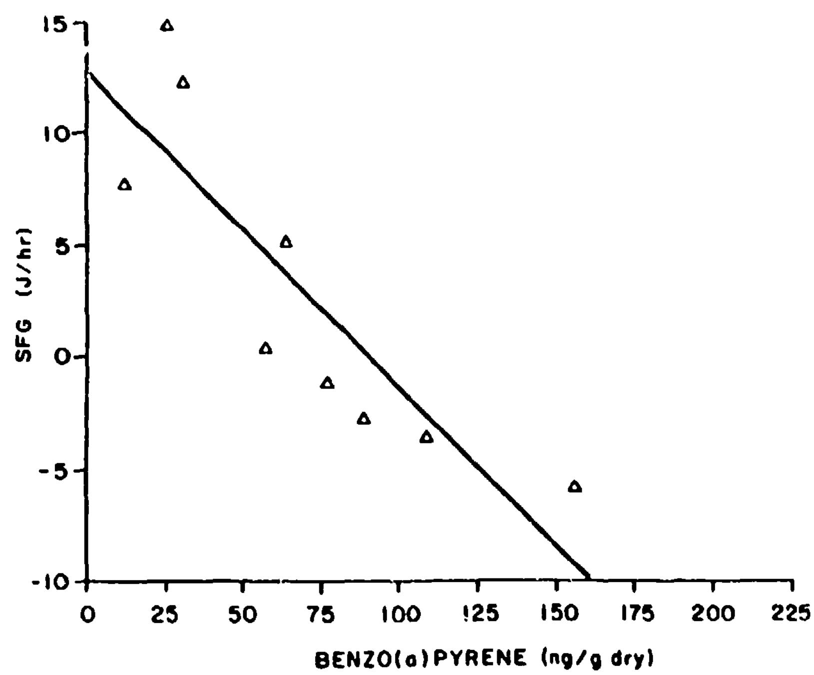


b. Sum of 178 alkyl homologs

Figure 18. Relationship between the SFG of *M. edulis* and the tissue residue concentrations of phenanthrene and the sum of the 178 alkyl homologs in the laboratory experiments. The presence of a regression line indicates a significant ($P < 0.05$) relationship between the two variables

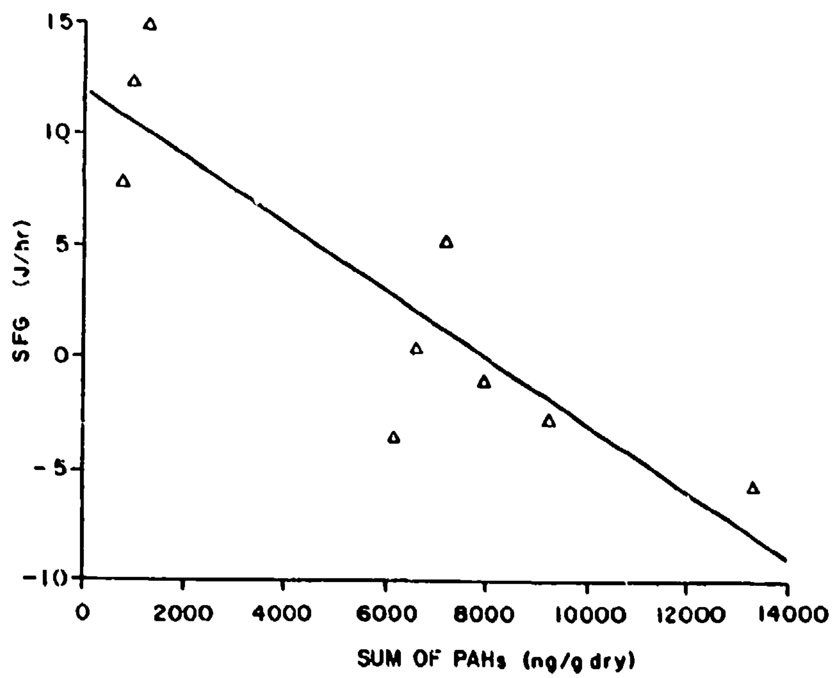


a. Fluoranthene

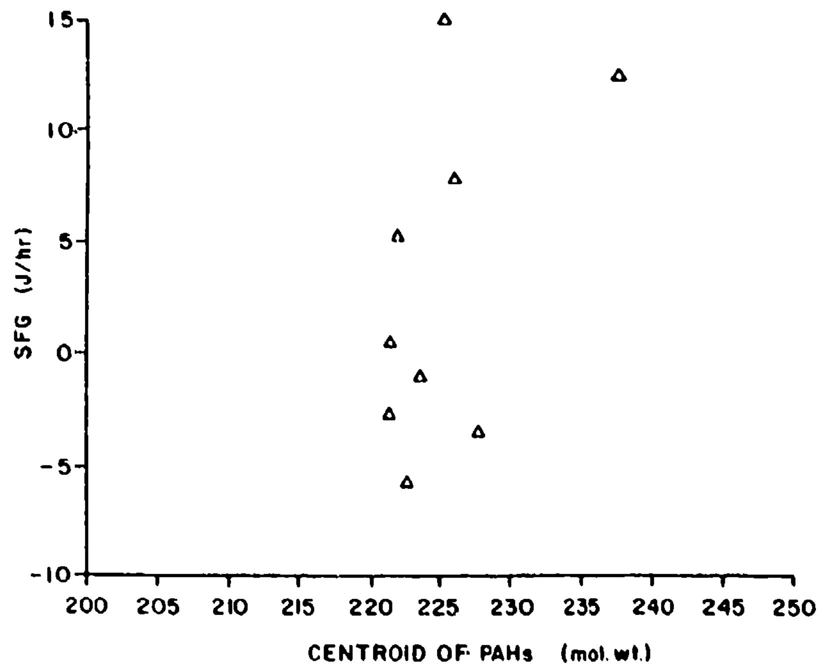


b. Benzo(a)pyrene

Figure 19. Relationship between the SFG of *M. edulis* and the tissue residue concentrations of fluoranthene and benzo(a)pyrene in the laboratory experiments. The presence of a regression line indicates a significant ($P < 0.05$) relationship between the two variables

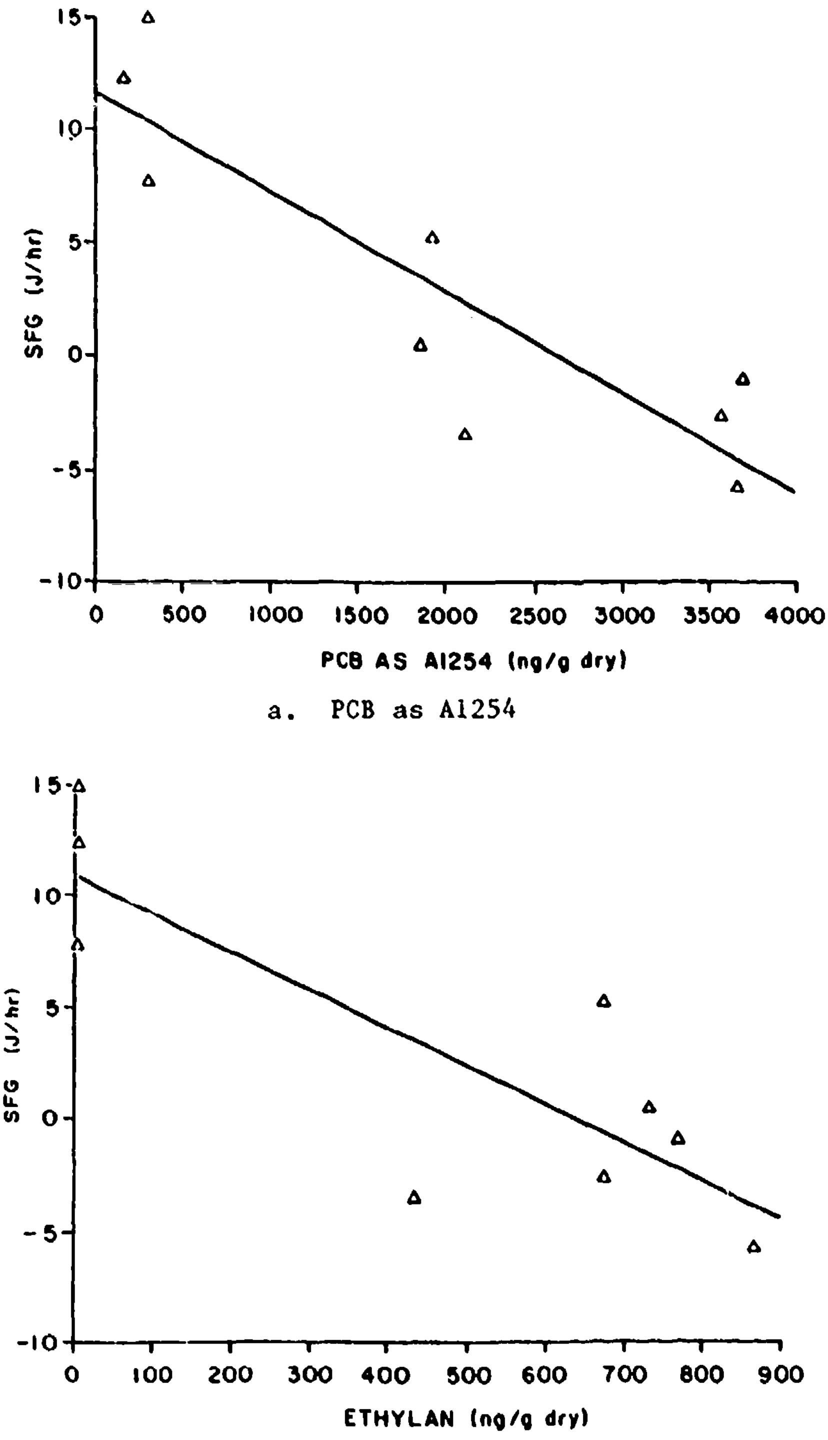


a. SUM of PAHs



b. CENT of PAHs

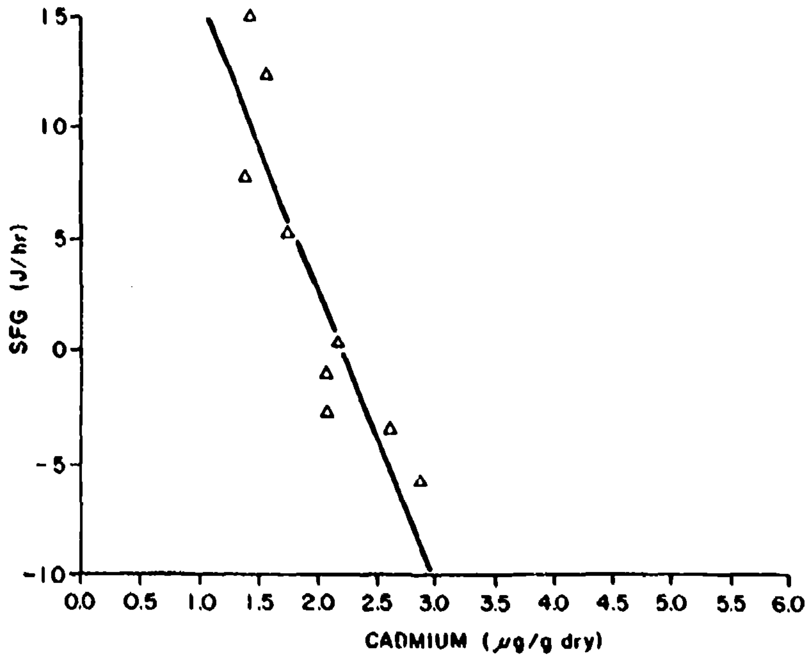
Figure 20. Relationship between the SFG of *M. edulis* and the summary statistics, SUM and CENT, in the laboratory experiments. The presence of a regression line indicates a significant ($P < 0.05$) relationship between the two variables



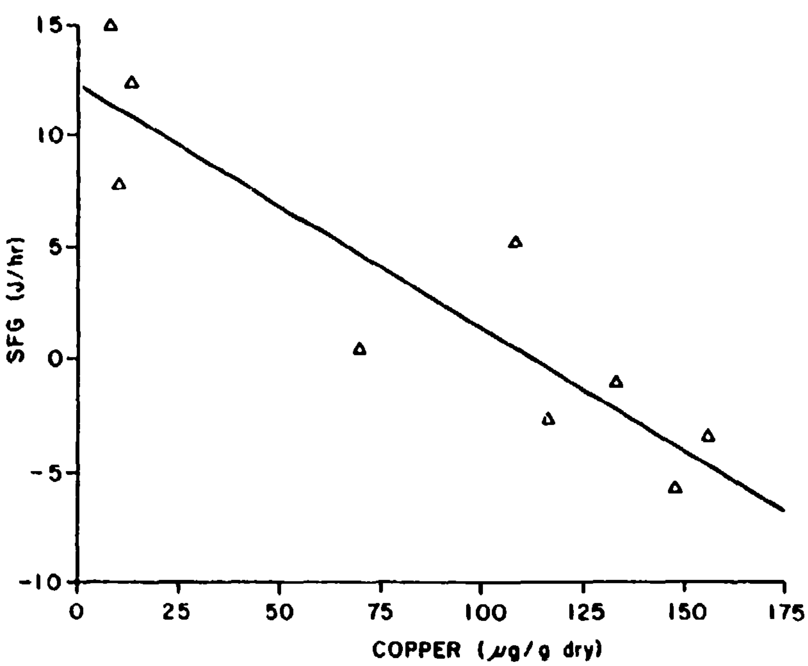
a. PCB as A1254

b. Ethylan

Figure 21. Relationship between the SFG of *M. edulis* and the tissue residue concentrations of PCB as A1254 and ethylan in the laboratory experiments. The presence of a regression line indicates a significant ($P < 0.05$) relationship between the two variables

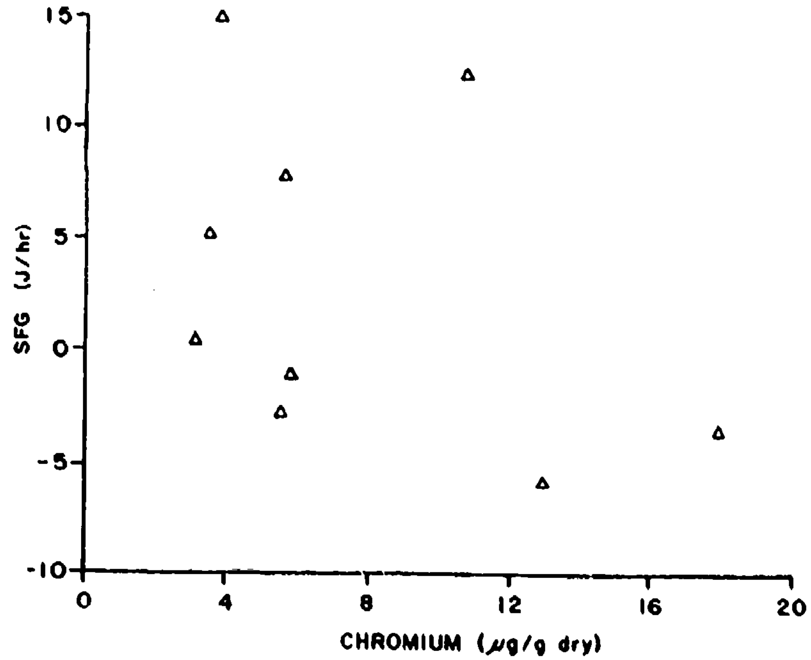


a. Cadmium

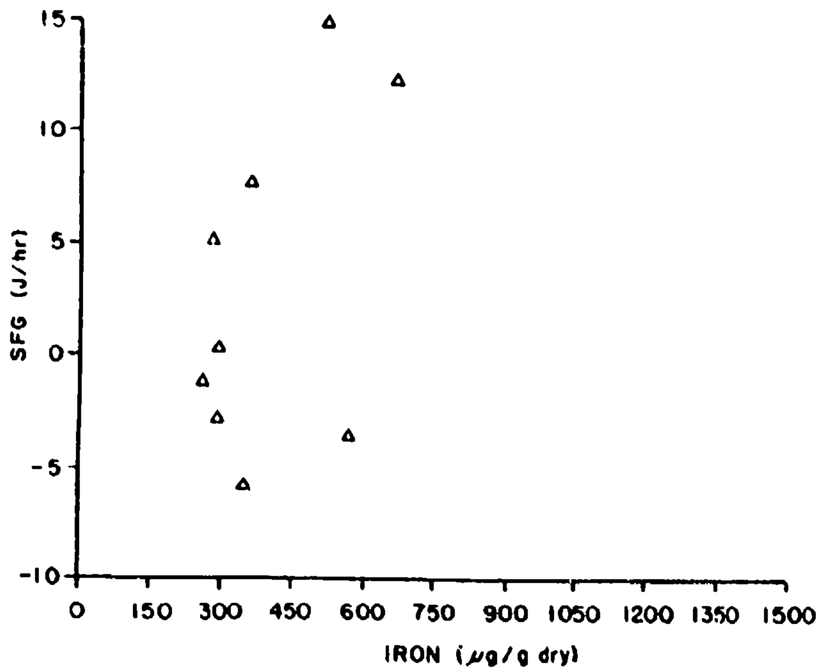


b. Copper

Figure 22. Relationship between the SFG of *M. edulis* and the tissue residue concentrations of cadmium and copper in the laboratory experiments. The presence of a regression line indicates a significant ($P < 0.05$) relationship between the two variables



a. Chromium



b. Iron

Figure 23. Relationship between the SFG of *M. edulis* and the tissue residue concentrations of chromium and iron in the laboratory experiments. The presence of a regression line indicates a significant ($P < 0.05$) relationship between the two variables

were selective as to which PAH compounds they accumulated (based on the CENT data), increased exposure levels of BRH material resulted in increased tissue residue concentrations of some PAH compounds (see paragraph 25).

96. There was also no apparent relationship between SFC and phenanthrene. This lower molecular weight compound was initially accumulated by mussels on Day 7 (Figure 18), after which the concentration decreased to a level similar in all three treatments. The fact that exposure remained constant while the phenanthrene tissue concentrations decreased over time may indicate the ability of the mussel to depurate or metabolize this compound.

Field

Exposure

97. Estimated from residues. The first method used to determine possible exposure conditions of *M. edulis* to BRH material in CLIS involved the use of laboratory-generated relationships between PCB tissue residues and BRH exposures. There are several assumptions inherent in this process: mussels provided an integrated measure of exposure during each field deployment; mussels were at equilibrium with background BRH levels in the water column; and PCBs are a good chemical marker for BRH material. Based on the results of the laboratory experiments, each of these assumptions seems reasonable.

98. The predicted exposures for each station and collection date demonstrate several spatial and temporal trends (Table 15). Spatially, the data indicate a trend toward greater exposure near the CNTR station immediately following disposal. This is evidenced by the elevated exposures at T = 0 (100E > REFS) and T + 2 (400E > 1000E > REFS) towards the disposal mound. This pattern disappeared by T + 8, where exposures were nearly the same at the CNTR, 400E, and 1000E stations, with the REFS station being lower than the other three.

99. Temporally, the estimated BRH exposures decreased with increasing time from disposal. The maximum exposure occurred at the 400E station at T + 2. This value ranged between 1.4 and 0.8 mg/ℓ of BRH suspended sediment, depending on whether the background concentration at REFS was subtracted. By the next collection, T + 8, the maximum estimated exposure, also at 400E, decreased to between 0.7 and 0.3 mg/ℓ, half that of the previous collection.

Table 15

Predicted BRH Suspended Material Sediment Exposure (mg/ℓ)

Required to Achieve the Measured Tissue Residue

Values of Mussels Deployed in CLIS*

Collection Cruise	Station	Estimated Exposure Range High Value	Estimated Exposure Range Low Value
T - 04	CNTR	0.37	0.00
	400E	0.26	0.00
	1000E	0.38	0.00
	REFS	0.38	0.00
T = 0	1000E	1.04	0.56
	REFS	0.49	
T + 2	400E	1.39	0.79
	1000E	0.98	0.38
	REFS	0.60	
T + 8	CNTR	0.67	0.21
	400E	0.71	0.25
	1000E	0.60	0.14
	REFS	0.46	
T + 12	CNTR	0.61	0.06
	400E	0.64	0.09
	1000E	0.53	0.00
	REFS	0.55	
T + 15	CNTR	0.84	0.31
	400E	0.61	0.08
	1000E	0.53	
T + 21	CNTR	0.52	0.12
	400E	0.66	0.26
	1000E	0.55	0.15
	REFS	0.40	
T + 27	400E	0.52	0.09
	1000E	0.37	0.00
	REFS	0.43	

(Continued)

* Each estimate was calculated based on laboratory-generated PCB residue-exposure concentration relationships. The high value was determined from the actual mussel tissue residue concentration while the low estimate was calculated after the REFS PCB residue was subtracted from the other stations during that collection period.

Table 15 (Concluded)

Collection Cruise	Station	Estimated Exposure Range High Value	Estimated Exposure Range Low Value
T + 43	CNTR	0.33	0.06
	400E	0.31	0.04
	REFS	0.27	
T + 55	400E	0.52	0.00
	1000E	0.42	0.00
	1000E	0.47	0.00
	REFS	0.53	
T + 116	CNTR	0.30	0.00
	400E	0.34	0.00
	1000E	0.43	0.01
	REFS	0.42	

Subsequent collections indicated a continued decrease to levels similar to those at the REFS station by T + 12.

100. Exposures estimated from water chemistry data. In addition to the estimates of BRH exposure based on mussel PCB tissue residues, a second estimate was made using PCB and copper concentrations in whole water samples taken postdisposal. The data indicate spatial and temporal trends similar to those obtained from the tissue residue estimates (Table 16).

101. Spatially, the sample collected on 7 June 1983 showed the highest BRH estimate (based on copper) at the CNTR station, followed by lower concentrations at 400E and 1000E stations, and the lowest levels at REFS. The estimate based on PCB concentrations indicated the CNTR station was elevated compared to the REFS station. The same pattern was observed in both the copper and PCB estimate for 21 July 1983 sample. A decreasing concentration of BRH material was estimated moving away from the CNTR of the disposal mound.

102. On a temporal scale, the BRH concentrations (copper data) decreased by about half from June to July (high estimate). After this collection, however, the copper-based BRH estimates fluctuated over time, with the December 1983 and June 1984 values higher than the September 1983 concentrations. This pattern over time may be more reflective of CLIS than of actual BRH levels because these estimates (high value) were based on t absolute copper levels at each location. Inspection of the low estimate indicated a more distinct pattern over the same time period. The BRH levels were highest immediately after the disposal operation (June 1983) and generally decreased with increasing time. The low estimate provided here is more a measure of relative difference between the stations, after background Long Island Sound concentrations are subtracted (REFS). When trying to discern temporal trends, this estimate may be more appropriate.

103. The pattern of BRH exposure based on PCB water concentrations was very similar to that of copper. The highest value was detected at the CNTR station in June 1983 and decreased both spatially and temporally with increasing time. In addition, the high estimates did not show the same variability over time that the copper data did. This may indicate that PCB concentrations in Long Island Sound were most constant over time, and thus BRH estimates based on PCB concentrations were less influenced by background fluctuations.

Tissue residues

104. The tissue residue levels for the mussels collected during the

Table 16

Predicted BRH Suspended Sediment Exposure (mg/ℓ) Based on PCB and Copper Whole Water Chemistry Data*

Date	Station	Estimate Using Cu High	Estimate Using Cu Low	Estimate Using PCB High	Estimate Using PCB Low
07 Jun 83	CNTR	1.30	0.71	1.05	0.69
	400E	1.12	0.53	--	--
	1000E	1.14	0.55	--	--
	REFS	0.59	0.00	0.36	0.00
21 Jul 83	CNTR	0.62	0.26	0.19	0.11
	400E	0.49	0.13	--	--
	1000E	0.41	0.05	--	--
	REFS	0.36	0.00	0.08	0.00
31 Aug 83	CNTR	--	--	0.17	0.07
	400E	--	--	0.21	0.11
	1000E	--	--	0.16	0.06
	REFS	--	--	0.10	0.00
02 Sep 83	CNTR	--	--	--	--
	400E	0.72	0.22	--	--
	1000E	--	--	--	--
	REFS	0.50	0.00	--	--
05 Dec 83	CNTR	--	--	0.05	0.00
	400E	1.13	0.37	0.08	0.00
	1000E	--	--	0.09	0.00
	REFS	0.76	0.00	0.09	0.00
06 Jun 84	CNTR	--	--	--	--
	400E	1.00	0.09	--	--
	1000E	--	--	--	--
	REFS	0.91	0.00	--	--

* Each estimate was calculated through division of the concentration of PCB or Cu present in the field by the concentration of that material present in the BRH barrel material (6,910 ng/g and 2,900 μg/g for PCB and copper, respectively). The high value was determined from the actual whole water concentration while the low estimate was calculated after the REFS values were subtracted from the other stations during that collection period.

course of the FVP study are presented graphically in Figures 24-29 for each of the 12 selected organic, inorganic, and summary statistic chemical contaminants. The raw data summarized in these figures are given in Appendix A.

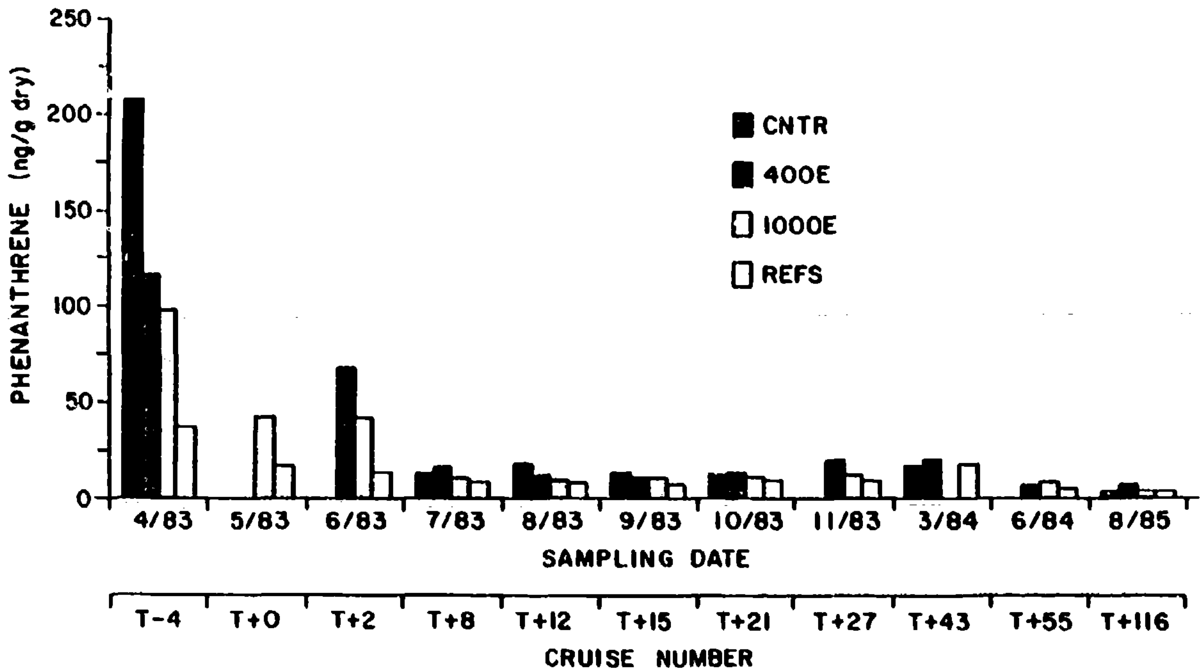
105. Temporally, the PCB, ethylan, and PAH residues increased during the disposal operation. After completion of the disposal, tissue residues decreased to concentrations similar to those from predisposal deployments. The summary statistic, SUM, reflected the same pattern as most of the PAH compounds.

106. A consistent pattern emerged when the spatial component of the organic residue data was considered within a sampling date. *Mytilus edulis* were deployed only twice during the actual disposal operation, at T + 0 and T + 2. For the T + 0 collection, only the 1000E and REFS stations were recovered. The tissue residue concentration for each organic compound was uniformly higher at the 1000E station than REFS. The T + 2 collection included data from three stations--400E, 1000E, and at REFS. Once again, a consistent pattern is seen in the residue data with mussels at 400E exhibiting the highest concentrations for each compound, followed by the 1000E and REFS stations. After the completion of disposal, the differences in residue concentrations decreased dramatically between stations.

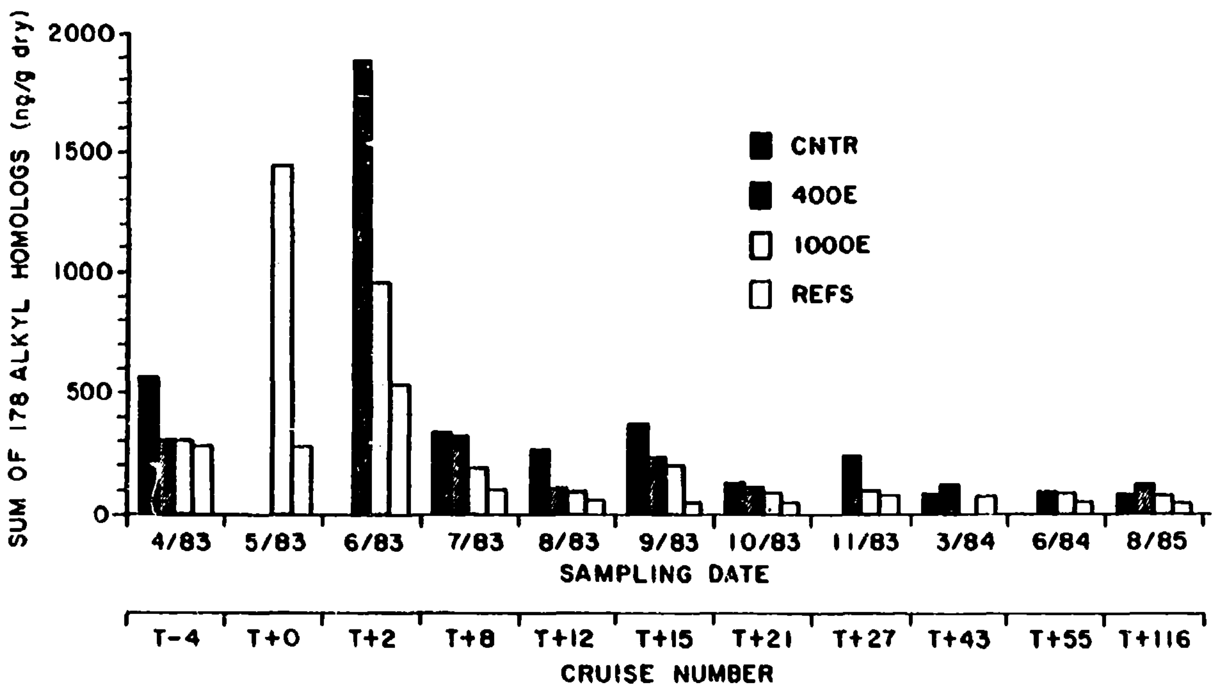
107. The tissue residue data for metals did not provide as clear a picture of the disposal operation as the organic residues. In general, metal residue concentrations increased slightly during the disposal operation, after which they decreased to levels well below those present during the predisposal collection (T - 4). Metal concentrations were elevated in *M. edulis* collected in October and November 1983 (T + 21, T + 27), well above those present even during the disposal operation (T + 0, T + 2). These two samples consisted of organisms that had been deployed at the FVP site for 7 months and 3 months, respectively. One possible explanation for elevated metals residues may be that mussels require a longer period of time to reach steady state with respect to metal concentrations. Comparing the organic and metal residue data from the field suggests that organic tissue residues present a better picture of the disposal operation at the FVP disposal site.

Scope for growth effects

108. The results of the physiological measurements are summarized in Table 17. Predisposal data, T - 4, indicated no differences among the physiological responses or the SFG index from each station (Table 17).

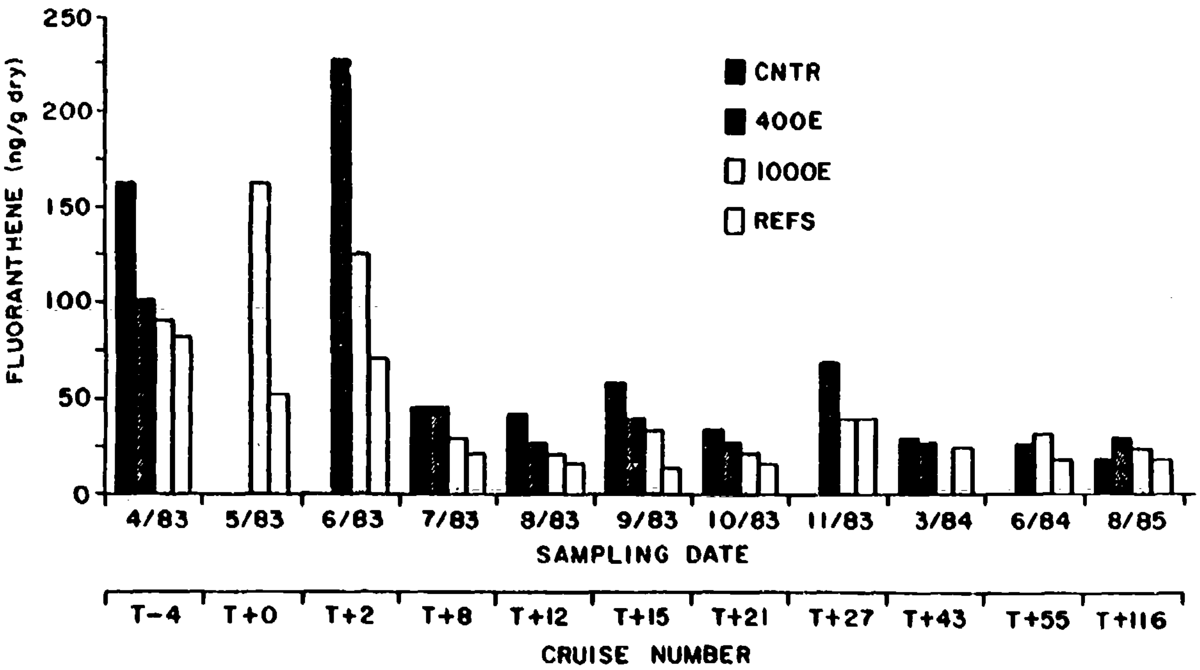


a. Phenanthrene

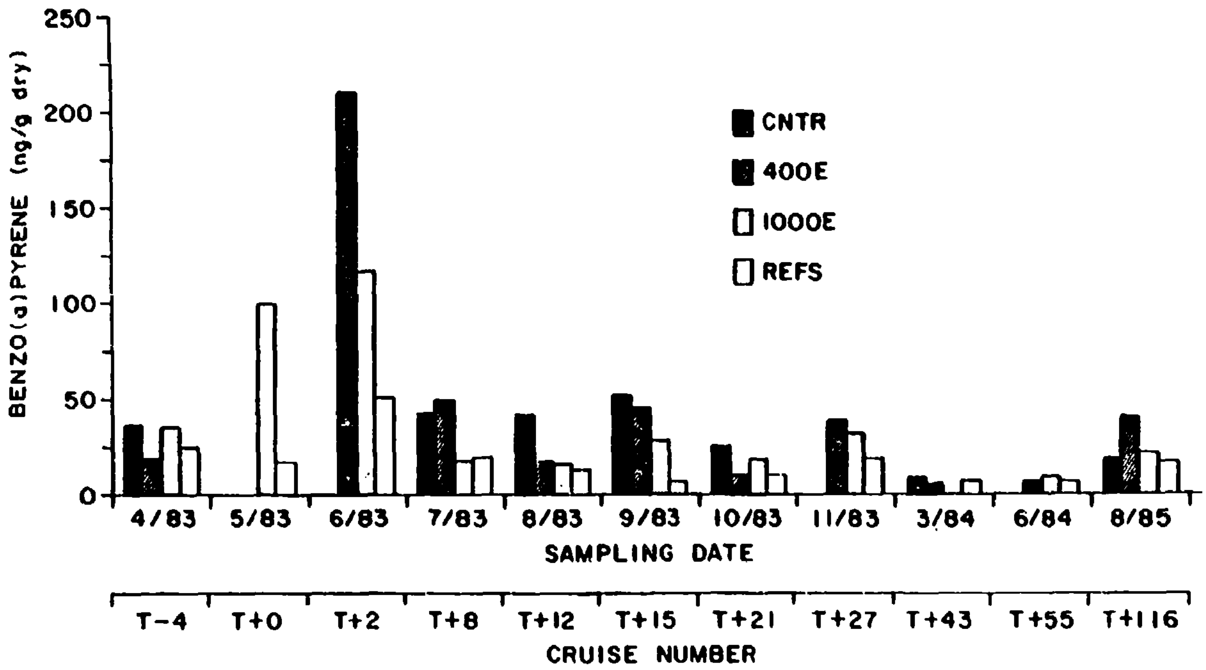


b. 178 alkyl homologs

Figure 24. Concentrations of phenanthrene and the 178 alkyl homologs in the tissues of *M. edulis* exposed at the specified FVP stations and sampling dates

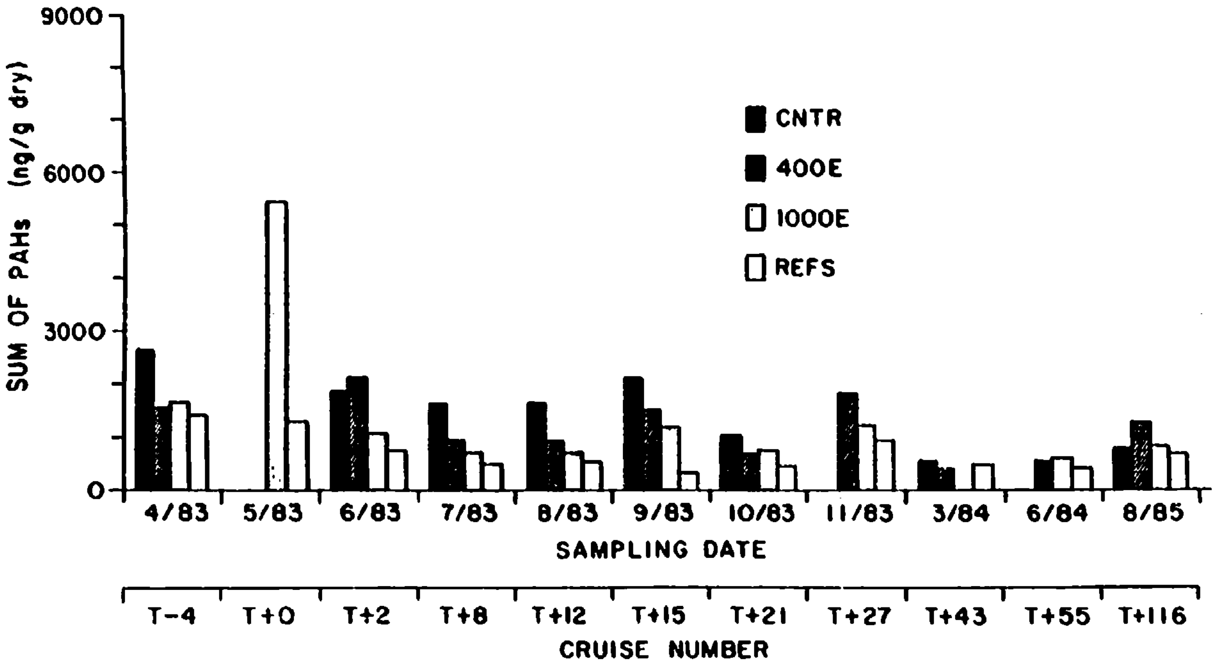


a. Fluoranthene

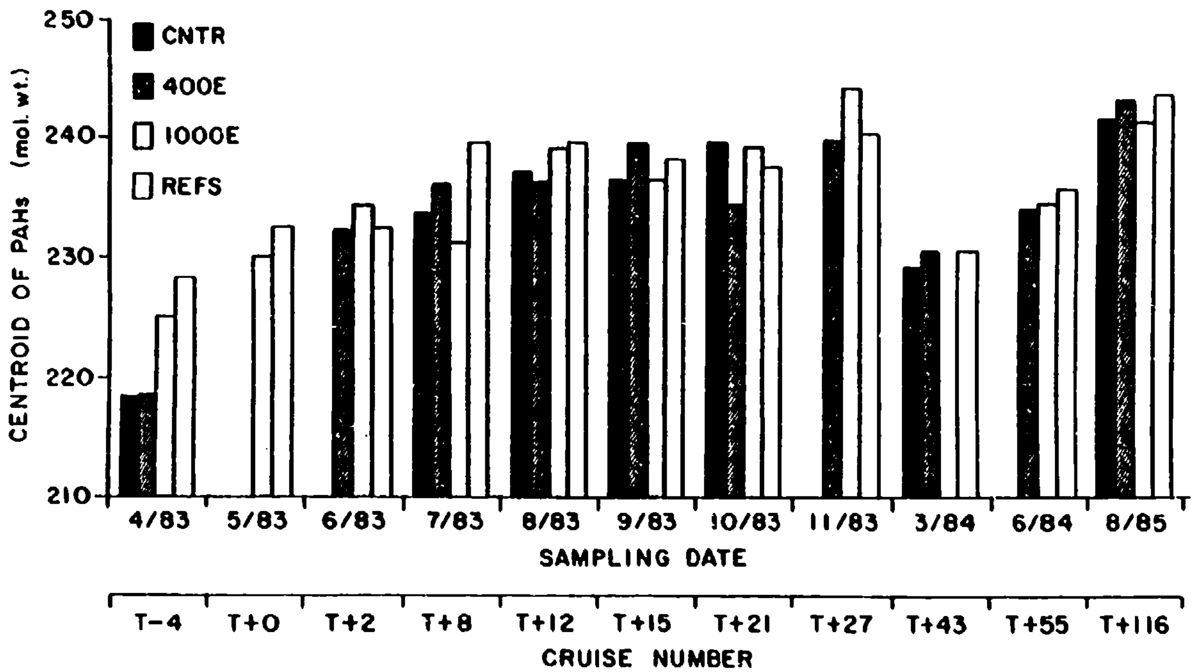


b. Benzo(a)pyrene

Figure 25. Concentrations of fluoranthene and benzo(a)pyrene in the tissues of *M. edulis* exposed at the specified FVP stations and sampling dates

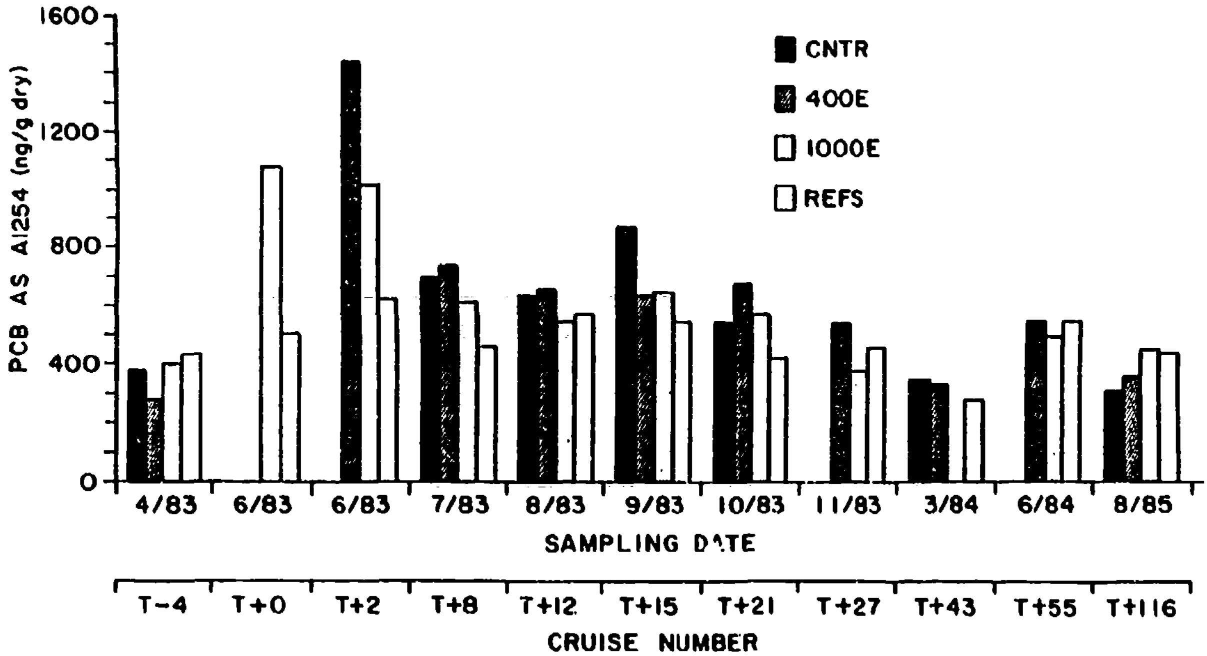


a. SUM of PAHS

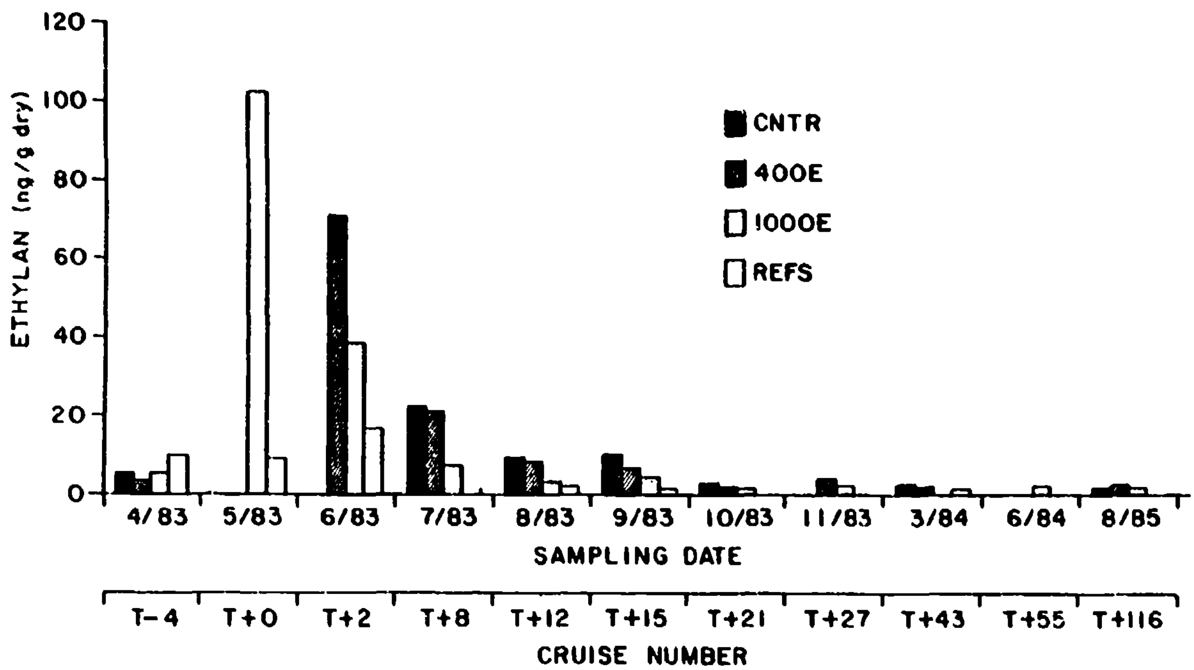


b. CENT of PAHs

Figure 26. Concentrations of the SUM of the PAHs and CENT in the tissues of *M. edulis* exposed at the specified FVP stations and sampling dates

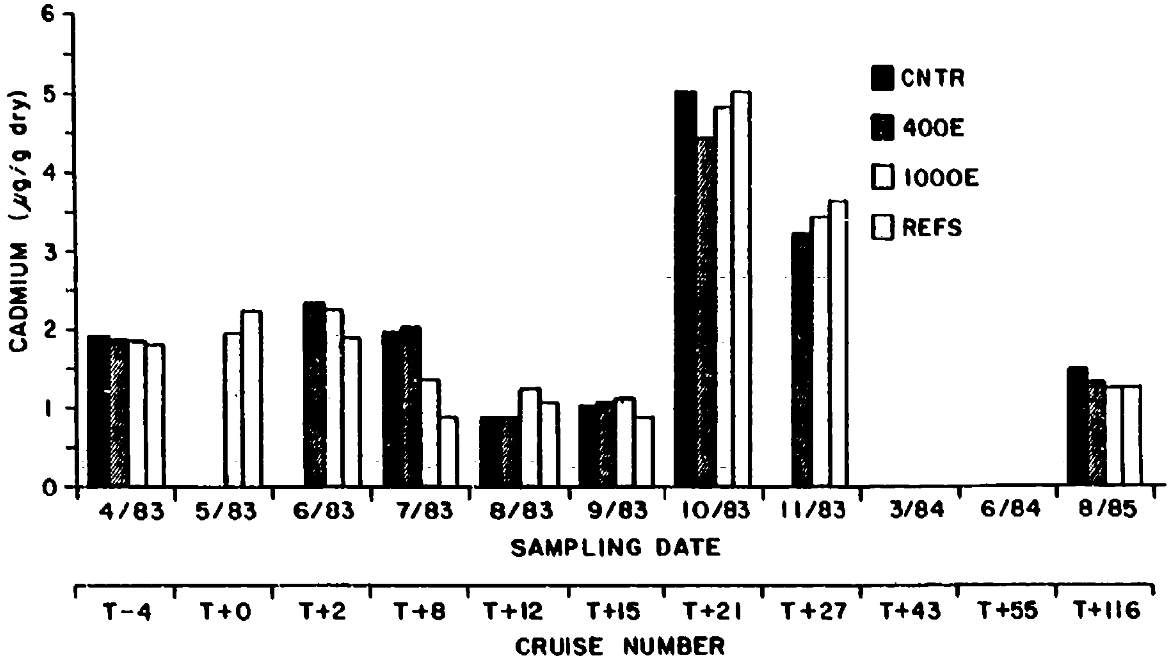


a. PCBs as A1254

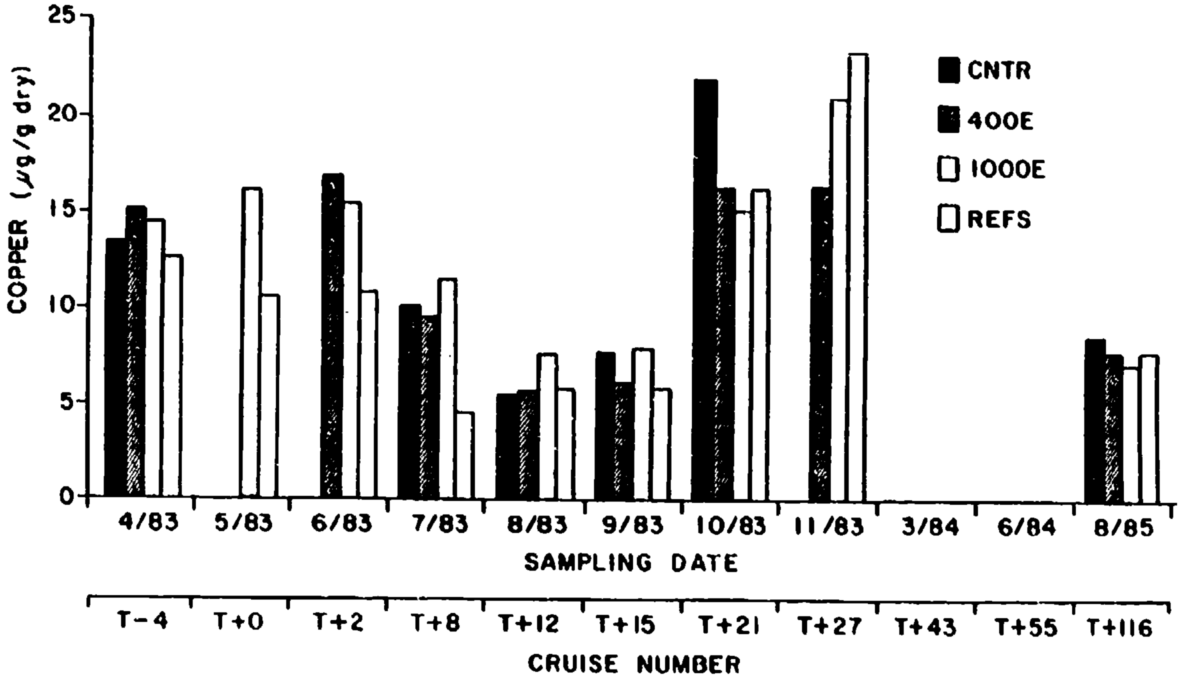


b. Ethylan

Figure 27. Concentrations of PCBs as A1254 and ethylan in the tissues of *M. edulis* exposed at the specified FVP stations and sampling dates

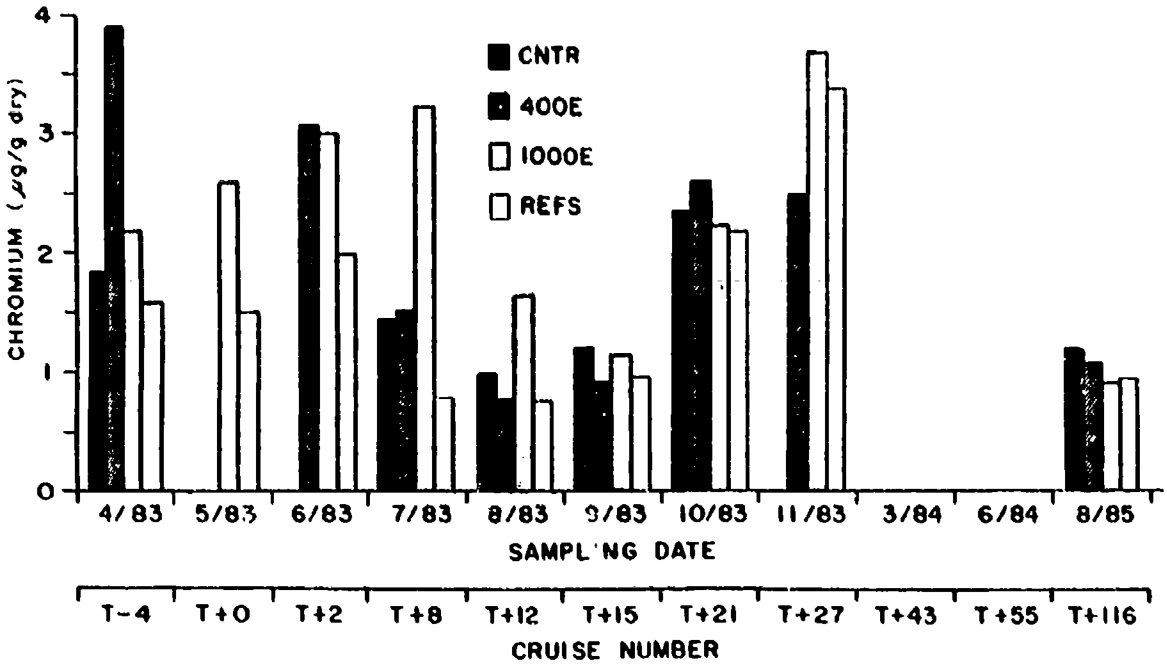


a. Cadmium

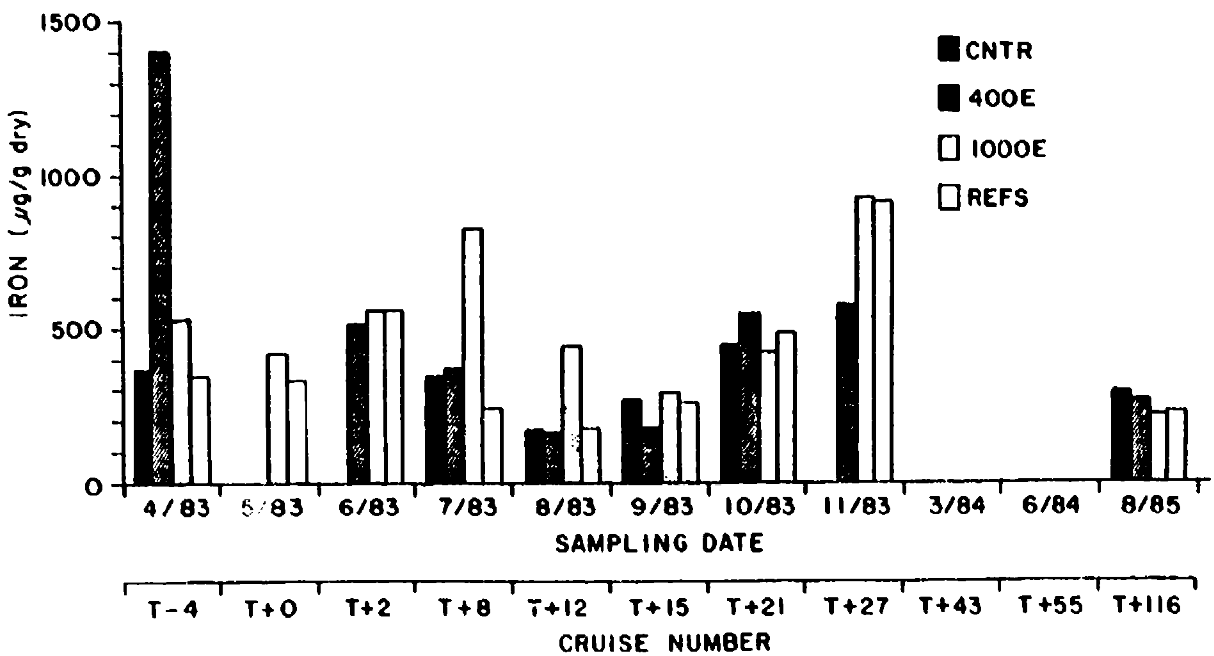


b. Copper

Figure 28. Concentrations of cadmium and copper in the tissues of *M. edulis* exposed at the specified FVP stations and sampling dates



a. Chromium



b. Iron

Figure 29. Concentrations of chromium and iron in the tissues of *M. edulis* exposed at the specified FVP stations and sampling dates

Table 17

Mean Values* (N = 10) of the Physiological Parameters Measured on Mussels from CLIS Collections with the Mean SFG Value Calculated for Station

Station	Clearance Rate ℓ/hr	Absorption Efficiency percent	Respiration Rate ml O_2/hr	Ammonia Excretion µg NH_4/hr	Scope for Growth J/hr
			T - 4, 22 Apr 83		
CNTR	2.35 (0.31)	94 (0.4)	0.61 (0.03)	23.08 (2.08)	3.33 (1.53)
400E	2.81 (0.25)	94 (0.5)	0.65 (0.02)	26.64 (1.28)	4.63 (1.45)
1000E	2.33 (0.21)	96 (0.4)	0.67 (0.04)	31.11 (4.00	2.31 (1.20)
REFS	2.20 (0.34)	95 (0.3)	0.66 (0.06)	21.51 (3.07)	1.36 (1.47)
			T + 0, 24 May 83		
1000E	4.88 (0.64)	92 (0.7)	0.72 (0.06)	16.51 (2.16)	9.46 (2.16)
REFS	5.05 (0.62)	91 (0.9)	0.73 (0.04)	13.99 (1.87)	8.87 (1.71)
			T + 2, 07 Jun 83		
400E	6.32 (0.54)	95 (0.4)	0.87 (0.04)	8.78 (1.26)	11.21 (1.76)
1000E	5.01 (0.58)	96 (0.7)	0.90 (0.05)	7.62 (2.17)	6.50 (1.38)
			T + 8, 13 Jul 83		
CNTR	3.53 (0.36)	88 (0.6)	0.95 (0.07)	20.19 (3.79)	-0.11 (1.18)
400E	4.63 (0.56)	89 (0.7)	0.84 (0.04)	21.81 (1.69)	5.04 (1.51)
1000E	5.69 (0.58)	83 (0.9)	0.91 (0.08)	19.80 (3.28)	6.30 (1.02)
REFS	5.39 (0.66)	85 (0.7)	0.80 (0.02)	14.08 (1.90)	6.85 (2.37)
			T + 12, 10 Aug 83		
CNTR	3.48 (0.31)	91 (1.1)	0.92 (0.16)	16.73 (3.28)	0.93 (1.47)
400E	4.02 (0.68)	94 (0.8)	0.95 (0.23)	14.40 (3.32)	2.73 (2.37)
1000E	3.43 (0.51)	96 (0.4)	0.97 (0.18)	40.35 (4.45)	0.18 (2.22)
REFS	3.65 (0.54)	96 (0.2)	0.88 (0.21)	34.19 (3.88)	2.97 (1.95)
			T + 15, 06 Sep 83)		
CNTR	3.78 (0.56)	92 (2.7)	0.77 (0.09)	33.85 (8.35)	4.70 (1.75)
400E	3.64 (0.44)	88 (4.7)	0.76 (0.04)	18.83 (2.91)	3.94 (1.74)
1000E	3.41 (0.57)	95 (4.8)	0.76 (0.06)	21.46 (2.92)	4.86 (2.77)
REFS	3.31 (0.35)	85 (4.6)	0.82 (0.06)	25.40 (1.88)	1.47 (1.74)

(Continued)

Note: Missing values during a collection period indicate the station was lost.

* Values in parentheses are standard errors.

Table 17 (Concluded)

Station	Clearance Rate ℓ/hr	Absorption Efficiency percent	Respiration Rate ml O_2/hr	Ammonia Excretion µg NH_4/hr	Scope for Growth J/hr
			T + 21, 18 Oct 83		
CNTR	1.91 (0.14)	91 (0.6)	0.74 (0.05)	27.18 (2.53)	-1.38 (0.98)
400E	1.69 (0.28)	89 (2.7)	0.66 (0.06)	28.35 (3.81)	-2.49 (1.81)
1000E	1.96 (0.28)	88 (0.6)	0.59 (0.04)	24.94 (1.75)	0.29 (1.76)
REFS	2.99 (0.43)	88 (1.5)	0.63 (0.05)	25.83 (2.78)	3.75 (1.54)
			T + 27, 29 Nov 83		
400E	1.78 (0.31)	96 (0.3)	0.42 (0.04)	24.65 (1.30)	4.53 (2.10)
1000E	2.06 (0.37)	96 (0.4)	0.52 (0.05)	26.39 (2.35)	3.08 (1.74)
REFS	1.63 (0.45)	95 (0.5)	0.37 (0.05)	23.47 (2.59)	2.71 (2.92)
			T + 43, 20 Mar 84		
CNTR	0.11 (0.05)	76 (6.1)	0.42 (0.03)	33.94 (2.50)	-8.50 (0.55)
400E	1.17 (0.19)	94 (0.6)	0.46 (0.03)	23.56 (2.50)	-0.54 (1.42)
REFS	1.58 (0.27)	95 (0.4)	0.55 (0.09)	46.85 (9.62)	0.40 (1.20)
			T + 55, 05 Jun 84		
400E	3.77 (0.76)	89 (0.4)	0.73 (0.05)	22.94 (2.88)	5.53 (2.61)
1000E	4.37 (0.57)	79 (1.7)	0.74 (0.07)	9.18 (1.33)	3.00 (2.31)
REFS	4.49 (0.40)	89 (0.7)	0.92 (0.05)	24.30 (2.89)	3.87 (1.31)
			T + 74, 17 Oct 84		
400E	2.52 (0.41)	76 (4.3)	0.48 (0.05)	23.98 (2.58)	1.65 (1.48)
1000E	3.55 (0.64)	75 (4.2)	0.54 (0.03)	37.31 (2.45)	5.27 (2.43)
REFS	3.55 (0.63)	77 (2.4)	0.51 (0.04)	23.31 (3.39)	6.29 (1.94)
			T + 116, 14 Aug 85		
CNTR	0.81 (0.12)	80 (1.0)	0.49 (0.03)	18.39 (2.81)	-4.24 (0.85)
400E	1.18 (0.19)	80 (0.5)	0.54 (0.04)	13.65 (2.65)	-3.01 (1.15)
1000E	1.39 (0.37)	78 (0.7)	0.68 (0.07)	14.32 (3.43)	-4.65 (1.55)
REFS	0.73 (0.08)	82 (1.6)	0.57 (0.06)	19.86 (3.78)	-6.02 (1.27)

109. Mussels collected at T + 0 and T + 2 had been deployed at the CLIS site for 1 month and 6 weeks, respectively. Differences in ammonia excretion rates were observed. However, the other physiological variables, as well as SFG values, were not different among stations. Loss of the 400E and REFS stations for the T + 0 and T + 2 collections, respectively, was unfortunate. This did not permit a comparison between the stations that presumably were most impacted (400E) and least impacted (REFS) during this time period.

110. A reduction in mean clearance rate was noted at the CNTR station compared with the rates at the 1000E and REFS stations during the T + 8 collection (Table 17), resulting in a subsequent reduction in the mean SFG of the CNTR mussels.

111. The SFG differences observed at the T + 8 collection were not present 1 month later in the T + 12 collection (Table 17). Mussels deployed for this monthly period exhibited higher mean ammonia excretion rates at the 1000E and REFS stations; however, there were no corresponding differences in the mean SFG index between stations.

112. No differences were observed among either the individual physiological parameters or the SFG index at the four stations at the T + 15 collection (Table 17).

113. The physiological measurements of the mussels collected at T + 21, after a 6-month deployment (Table 17), indicated a difference in clearance rates and SFG among stations. Mussels from the REFS station had a higher mean clearance rate than the mussels from the other three stations. The SFG data indicate a lower mean SFG at the CNTR and 400E stations compared with the REFS station, with the mean SFG of the 1000E mussels not different from the other three stations.

114. The T + 27 mussel collection was completed after a 3-month deployment at the FVP disposal site. An increase in the mean respiration rate was noted in the mussels collected from the 1000E station (Table 17). No differences were present in the mean SFG values among the stations.

115. The physiological data for the mussels collected at T + 43, after a 3-month deployment (Table 17), showed that mean clearance rate, mean absorption efficiency, and mean SFG were lower in mussels from the CNTR station compared with those of the mussels from the other two stations. The mean clearance rate of the CNTR mussels was almost zero. In addition, the mean absorption efficiency was lower than the other stations. Small differences in

absorption efficiency of 1 to 5 percent may not be biologically significant due to measurement variability. In this instance, however, the mean absorption efficiency of mussels from the CNTR station was almost 20 percent lower. Finally, the mean SFG response of the mussels from the CNTR station was lower than the mussels at the 400E and REFS stations.

116. The mussel collection at T + 55 represented a deployment period of 8 months (Table 17). There were no differences between the mean SFG values for any of these stations; however, mussels retrieved from the 1000E station exhibited lower mean absorption efficiencies and lower mean ammonia excretion rates than mussels from the 400E and REFS stations.

117. Mussels retrieved during the T + 74 collection had been deployed for a period of 4 months (Table 17). Mean ammonia excretion rates were higher in mussels from the 1000E station than those of the mussels from the 400E and REFS stations. There were no differences among the mean SFG values from the three stations.

118. The final collection from CLIS, T + 116, was completed in August 1985. Mussels were deployed for a period of 1 month to observe whether there were any long-term effects from the disposal of the BRH material. The data indicate no differences among stations for any of the physiological measurements (Table 17); however, the SFG values were low at all stations.

Residue-effects data

119. The relationship between SFG effects and tissue residues measured in the field mussels was not as clear as it was for the laboratory experiments. The SFG of a mussel is influenced by a variety of extrinsic (i.e., temperature) and intrinsic (i.e, gametogenesis) factors which varied naturally from collection to collection during the FVP. These normal seasonal changes in SFG preclude regressing all of the field SFG and tissue residue data together because they would conceal any relationship that might exist. Therefore, the field residue-effects data will be considered within discrete sampling periods, where mussels were presumably exposed to similar temperatures and should be in a similar stage of gametogenesis. As was stated at the beginning of this report, the purpose for using the SFG index was to determine whether relative sublethal effects could be measured between laboratory treatments or field stations. Consideration of field SFG-residue relationships within a sampling period is entirely consistent with this objective.

120. The use of SFG values within a sampling period reduced sample size

to a maximum of four, when all stations were recovered. Regression analysis with this sample size is not appropriate; therefore, the data are presented graphically to illustrate trends. It would be impractical to present graphs for all of the residue-effects data for each of the 12 sampling dates. PCBs were selected because of the good relationship previously described between residues of this compound and SFG in the laboratory experiments, as well as the relationship between BRH exposure concentration and mussel PCB residues in those same laboratory experiments. Therefore, only the field residue-effect data for PCBs and SFG will be presented here. A complete comparison of residue-effects data for the field collections can be made by comparing field SFG values (Table 17) with field mussel residues (Appendix A, Tables A3-A13).

121. The SFG-PCB tissue residue data are presented in Figure 30. Mussels collected predisposal showed a very narrow range of PCB residues with a corresponding small range in SFG values. The T = 0 collection indicated that mussels from 1000E exhibited elevated PCB residues compared with REFS; however, SFG values from both stations were similar. Mussels collected at T + 2 once again displayed higher residues closer to the disposal mound, 400E compared with 1000E, while SFG values were not that different from each other. Subsequent field collections indicated a reduction in PCB tissue residues from all stations. At T + 8, mussels from the CNTR station had a lower SFG value compared with those at the other three stations; however, there was no corresponding difference in PCB residues at this time. Mussels from the T + 12, T + 15, T + 21, T + 27, T + 55, and T + 116 collections exhibited a very small range of PCB tissue residues and a corresponding narrow range between SFG values at the four stations. The only exception was the T + 43 collection where a reduction in SFG at the CNTR station was not related to any differences in PCB residues among stations.

122. These data indicate that no distinct relationship was evident between SFG and tissue residues in the field. Several possible explanations will be put forth in the discussion; however, one fact will be mentioned here. The highest mussel PCB residue concentration in the field (T + 2, 400E) was less than the lowest tissue residue in the laboratory experiments (10 percent, Day 28). These data suggest that residue concentrations of field-exposed mussels may have been too low to elicit a SFG effect.

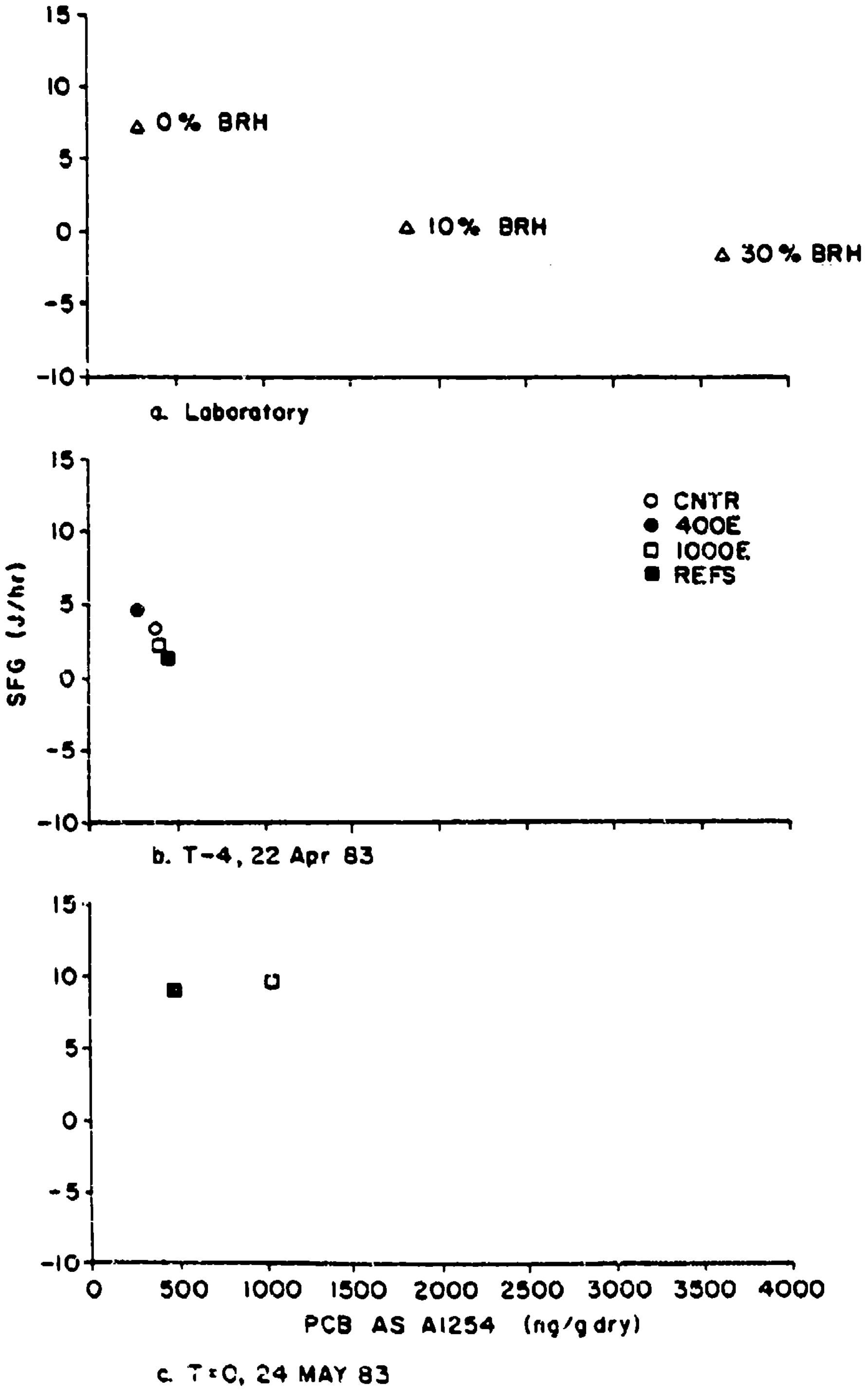


Figure 30. Relationship between the SFG of *M. edulis* and PCB tissue residue concentrations in laboratory and field-exposed animals. The laboratory data are presented to provide a perspective between the residue concentrations of laboratory and field-exposed mussels (Sheet 1 of 4)

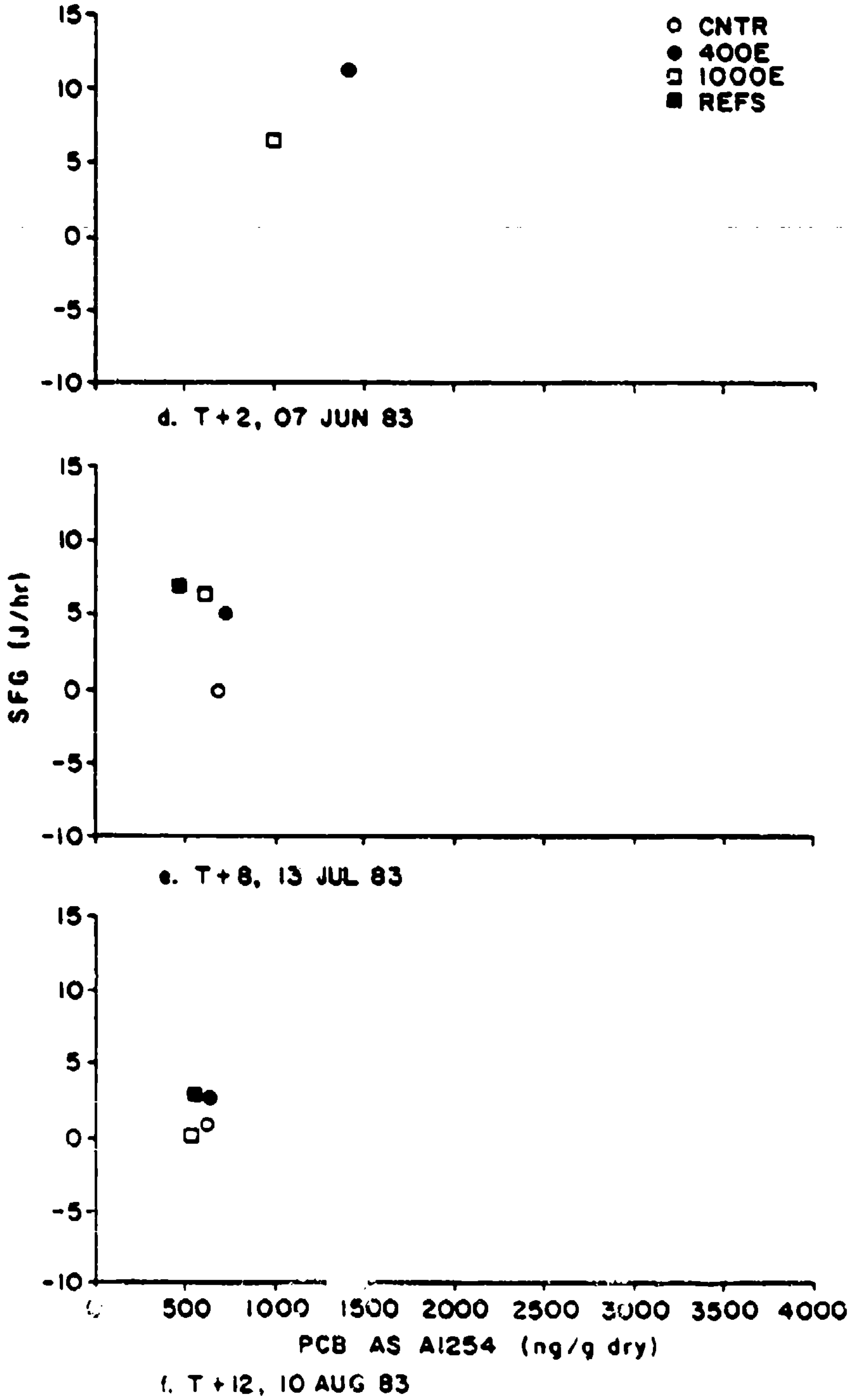
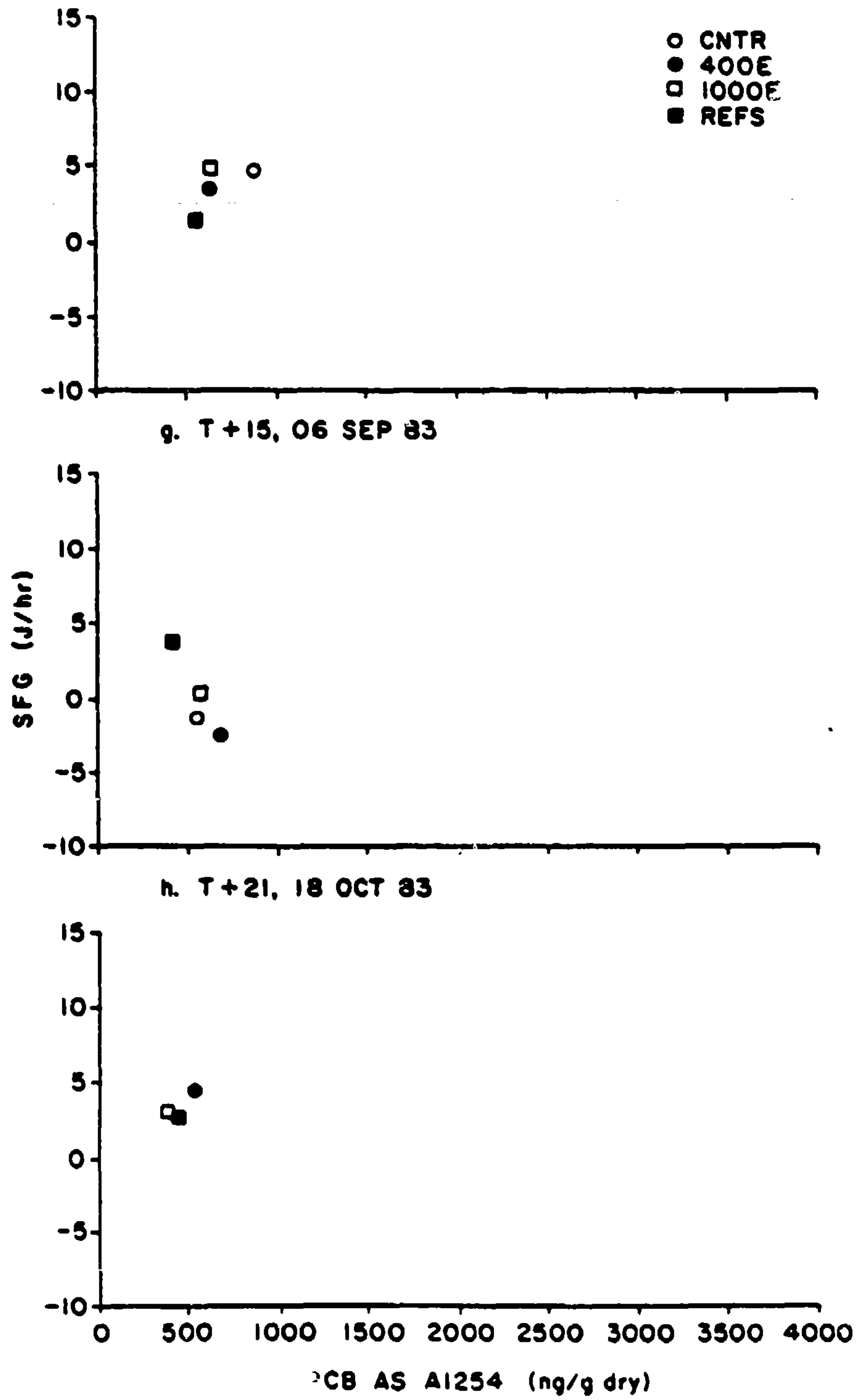


Figure 30. (Sheet 2 of 4)



i. T + 27, 29 NOV 83

Figure 30. (Sheet 3 of 4)

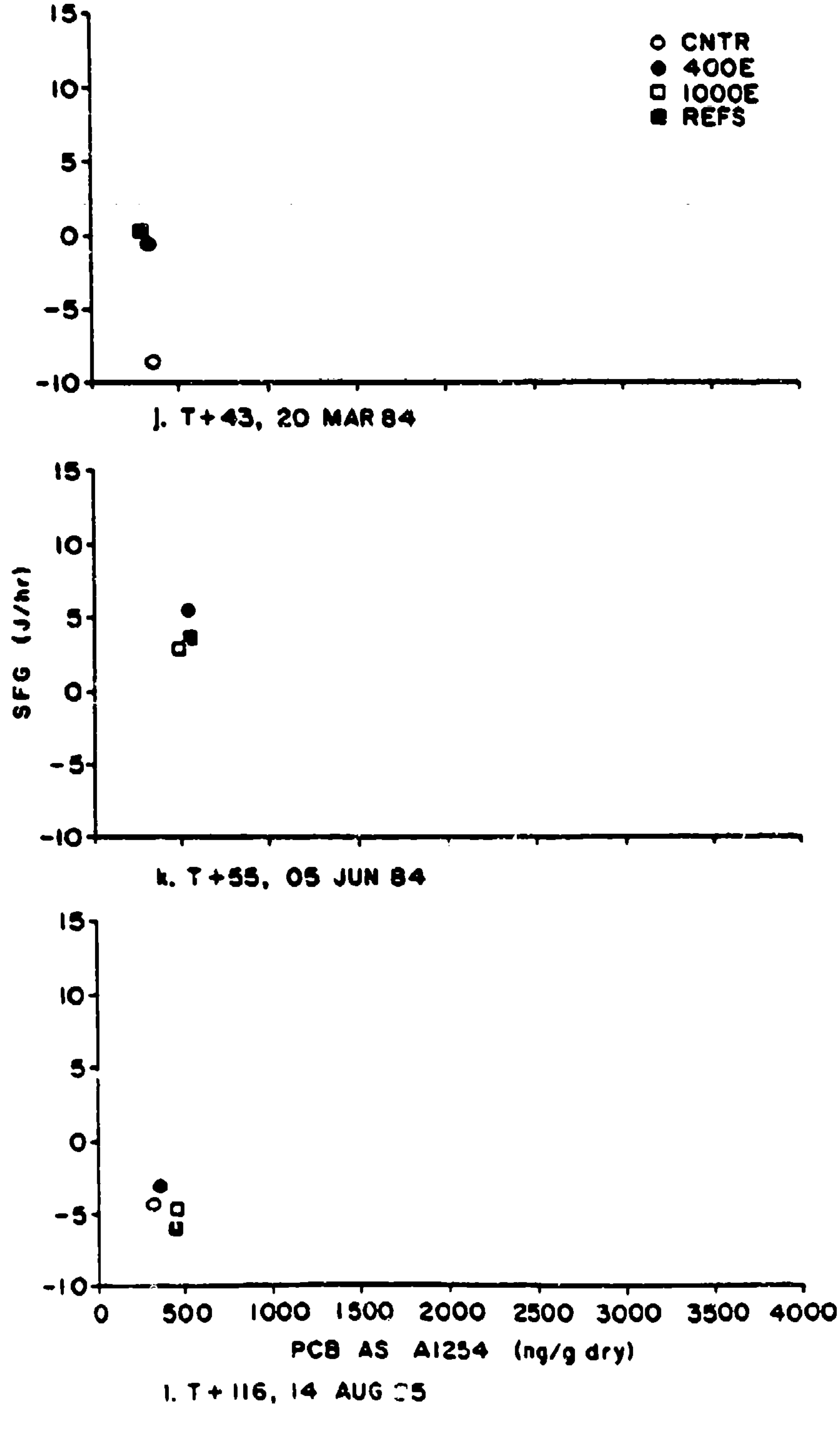


Figure 30. (Sheet 4 of 4)

Laboratory-to-Field Comparison

123. The laboratory-to-field comparison was completed in two parts and included both tissue residue and effects data. The approach taken was to first establish when exposure conditions were similar in the laboratory and the field by comparing laboratory and field residue data. Comparable tissue residues were interpreted as being indicative of comparable BRH exposures. The second step was to compare the SFG values of the laboratory- and field-exposed mussels with similar tissue residue concentrations.

Residues

124. Results of the cluster analysis suggested several general observations. First, the samples that were most similar included all the field residues collected after T + 2 and the laboratory 0-percent BRH exposures. This would indicate that mussels in the field received minimal exposure to BRH material after the initial disposal operation. Second, mussels collected predisposal (CNTR, 400E, 1000E) and those collected shortly after disposal (1000E at T = 0 and T + 2) were more similar to the other field samples than the laboratory samples. This would imply that even during disposal, BRH exposures at these stations were more similar to subsequent postdisposal field residues than to laboratory BRH exposures. Third, all of the laboratory residues were more similar to each other than to any of the field samples. This grouping would indicate that all laboratory exposures were very different from field exposures. Finally, mussel residues obtained from 400E at T + 2 were more similar to those of the laboratory-exposed mussels than to the other field exposures. This sample was the last to cluster, indicating that it was not very similar to any other samples; however, it was more closely related to the laboratory samples than to the field samples.

Effects

125. Analysis of the residue data suggested that the most valid comparison between laboratory and field SFG data would be between field samples, with the exception of 400E at T + 2, and laboratory mussels exposed to 0-percent BRH. Comparison of the SFG of mussels exposed to even 10-percent BRH in the laboratory to any mussels in the field would not be appropriate because the residues were dissimilar. Additionally, comparison of mussels collected from the field when environmental conditions were not similar to those in the laboratory would not be appropriate.

126. Laboratory experiments were conducted in the spring at water temperatures of 15° C. Comparable field conditions existed at T = 0 and T + 2 in the field. The SFG value of mussels exposed to 0-percent BRH in the lab for 28 days was 7.2 J/hr. The field-exposed mussels exhibited SFG values of 9.5, 8.9, and 6.5 J/hr, respectively, for the T = 0 (1000E, REFS) and T + 2 (1000E) collections. The SFG values were very similar, indicating that the relative physiological conditions of these mussels were the same when environmental conditions were similar. The only other collection that occurred in the spring when water temperatures were similar was at T + 55; however, these mussels had been deployed in CLIS for 8 months. The SFG values of these mussels, 3.8, 3.5, and 3.5 J/hr, were slightly lower than that in the 1-month laboratory exposure (7.7 J/hr).

127. Several generalizations are apparent from the comparison of laboratory and field results. The laboratory exposures indicated a good relationship between BRH exposure and residue concentrations in *M. edulis*. These data provide justification for assuming that lower residue concentrations in the field-exposed mussels were indicative of lower BRH exposures in the field. In fact, the highest field residues were less than the residues of the mussels exposed to the lowest BRH concentration (1.5 mg/ℓ) in the laboratory. Therefore, the data suggest that there was little or no overlap in laboratory and field BRH exposure concentrations. The resultant effects data indicated dramatic adverse effects on mussels even at the lowest laboratory exposure concentration whereas, in the field, few if any effects were attributable to BRH exposure.

PART IV: DISCUSSION

128. The objectives of this study were to: (a) investigate the residue-effect relationships in the mussel after independent laboratory and field BRH exposures and (b) field verify the laboratory results. The design of this study followed a logical progression from BRH exposure to tissue residue concentration to biological effects. The discussion will parallel this approach by establishing the exposure-residue and residue-effect relationships separately in the laboratory and in the field. Finally, a comparison of the laboratory and field results will be made.

Laboratory Experiments

129. There was a strong link between exposure to BRH sediment and subsequent tissue residues in *M. edulis*, as confirmed by the monitoring data collected during the laboratory experiments. In addition, the relationship between mussel tissue residues for several chemical contaminants demonstrated that compounds with higher molecular weights and stability, PCBs in particular, tracked the BRH exposure concentrations remarkably well. For example, tissue residue data indicated that mussels from the 30-percent BRH chamber exhibited twice the level of PCBs as those in the 10-percent BRH chamber. Corresponding monitoring data indicated that the actual delivered level of BRH sediment was 3.3 and 1.5 mg/ℓ, respectively, for those two chambers, indicating that PCBs were a good "marker" for exposure to BRH material. Because of this direct relationship, residue concentrations can be assumed to be indicative of exposure concentration for highly stable compounds such as PCB. This relationship is particularly important in the field where direct, continuous monitoring data of exposure conditions are difficult, if not impossible, to collect.

130. In addition to the strong exposure-residue relationship, the laboratory experiments indicated a good relationship between mussel residue concentrations and biological effects. The residue-effects data presented in this study were never intended to determine cause and effect. However, relationships between individual contaminants and biological effects present strong evidence that observed reductions in scope for growth, clearance rate, and shell growth were related to exposure to BRH material.

131. Mussel SFG values were reduced after exposure to BRH dredged material on Day 14 of the first experiment and both Days 14 and 28 of the second experiment. While there is no attempt to attribute observed decreases in SFG in the present experiments exclusively to any one contaminant in the BRH sediment, there is evidence to suggest that some of the contaminants are capable of causing the observed SFG reductions. Stickle et al. (1985) reported an inverse relationship between SFG of mussels and water-soluble fraction aromatic hydrocarbon concentrations. Widdows et al. (1982) have demonstrated reductions in the SFG of mussels exposed to the water-accommodated fraction of North Sea oil. Gilfillan (1975) reported a net decrease in carbon flux in *M. edulis* after exposure to crude oil extracts. Copper was also found to reduce the SFG of *M. edulis* (Moore et al. 1984).

132. In addition to the fact that BRH material affected SFG, the shape of the dose-response curve at Day 14 is also of interest (Figure 14). This relationship, best described by a curvilinear function, implies that exposure to some BRH concentration less than 3.3 mg/ℓ could have no effect on SFG in mussels, i.e., a possible "threshold" concentration of BRH material is required to cause adverse physiological effects. A similar trend was displayed by the Day 28 data where a relatively small SFG difference was found between mussels exposed to 1.5 mg/ℓ and 3.3 mg/ℓ (0.14 and -1.8 J/hr, respectively) compared with mussels exposed to no BRH material (7.16 J/hr).

133. Inspection of the individual physiological parameters indicated that reductions in SFG may be related exclusively to decreased clearance (feeding) rates. Absorption efficiencies, respiration rates, and ammonia excretion rates were not different among treatments at Day 14 or Day 28 (Tables 10-12). The impact of BRH suspended sediment on clearance rates was consistent in both experiments and almost identical to that between SFG and BRH levels (Figures 15-17). This type of response was observed by Stickle et al. (1985) and Widdows et al. (1982) in *M. edulis* after exposure to oil extracts. Gonzales et al. (1979) reported reduced clearance rates in *M. edulis* after exposure to No. 2 heating oil. Nelson, Black, and Phelps (1985) found lower clearance rates in mussels after exposure to anaerobic BRH dredged material. Reductions in clearance rate have been observed in other species as well. Gilfillan et al. (1976) reported a reduction in filtration in the soft-shelled clam *Mya arenaria* from areas exposed to oil spills. Stickle, Rice, and Moles (1984) found that reductions in the SFG of the gastropod *Thais lima*

after exposure to hydrocarbons were primarily due to reduced feeding rates.

134. Clearance rate measurements in the exposure system provide further evidence that BRH suspended sediment affected clearance rates. Measurements on Day 9 in the first experiment (Table 8) demonstrated a dramatic reduction in the clearance rates of mussels from the 50- and 100-percent BRH treatments. Measurements in the second experiment, on Days 7 and 16, indicated that exposure to as little as 1.5 mg/ℓ BRH for 7 days was sufficient to produce dramatic clearance rate reductions. There was the possibility, initially, that the reduced clearance rates in the exposure chambers represented a behavioral response (i.e., the mussels reduced their clearance rates in response to the BRH material). However, the SFG measurements were made in the absence of BRH material. Therefore, the differences observed in the clearance rates of mussels from the exposure system were not due to behavior. Rather, reductions in clearance rates were probably due to physiological impairment from the BRH material.

135. One explanation for the reduced clearance rates has been suggested by histopathological observations. Mussels from treatments containing BRH suspended sediment showed a loss of cilia from the gill filaments, while those exposed to REFS sediment alone were normal (Yevich et al. 1986). This information is consistent with clearance rate being the only parameter altered in the BRH treatments because the coordinated movement of these cilia is responsible for creating the currents necessary to move particles into and out of the mussel. The present study augments the evidence in the literature that clearance rates are particularly sensitive to pollutants of the type found in the BRH dredged material.

136. Shell growth data (Table 14 and Figures 15-17) supported the relative effects of BRH sediment shown by the SFG index. Actual growth observations could be explained on the basis of the SFG and clearance rate measurements completed over this same time period. Reduced clearance rates in the treatments containing BRH sediment resulted in decreased energy consumed by those mussels. These reductions occurred in as little as 7 to 9 days, evidenced by the data in Table 8. The adverse effect of the BRH sediment was observed in the physiological measurements on Day 14. Reduced SFG values indicated that relatively less energy was available for growth with increasing concentration of BRH, which is exactly what the growth data reflected. Exposure to BRH sediment for an additional 14 days produced continued low SFG

values in the 10- and 30-percent BRH treatments, and subsequent lower growth in these mussels over the second 14-day period.

137. Agreement between the SFG index and shell growth has been reported by Bayne and Worrall (1980) for mussels and Gilfillan and Vandermeulen (1978) in *M. arenaria*. The close correspondence between the SFG response and shell growth in the present study supports the use of standardized conditions to measure relative sublethal effects in mussels. The measurement of SFG under standardized conditions is sometimes criticized as being nonreflective of what occurred under field exposure conditions. The results of this study indicated that the relative differences in SFG values were indicative of actual changes in shell growth.

138. The similarity between SFG and actual growth raises the obvious question: why not measure shell growth alone and omit SFG altogether? The SFG index reflects the total energy available for both somatic growth and reproduction. Energy available for somatic growth can be further partitioned into either tissue growth or shell growth. Changes in shell length alone quantify only one component of the organisms' response to the environment, while the SFG index quantifies the total energetic response. The ultimate question in pollution biomonitoring is whether or not changes in biological effects measurements on individuals reflect changes in populations and communities. Reductions in SFG have been correlated with a decrease in reproductive output of individual mussels, and, therefore, may be indicative of broader ecological consequences as well (Bayne, Clark, and Moore 1981). Changes in shell growth alone, while important, may not provide as much information about possible ecological effects as SFG. Whenever possible, both SFG and actual growth should be measured concurrently to provide as much biological effects data as possible.

139. To summarize the laboratory studies, tissue residues in *M. edulis* were directly related to exposure concentration of BRH material. PCBs, in particular, were a good marker for BRH suspended sediment exposure concentration. A consistent, inverse relationship was observed between the SFG, clearance rate, and shell growth of mussels and increased exposure to BRH suspended sediment. Relative differences in SFG were supported by similar changes in shell growth. Based on these data, a BRH suspended sediment concentration equal to 1.5 mg/ℓ would be predicted to cause a similar effect in the field.

Field Experiments

140. The exposure-residue relationships generated in the field portion of this study were not as straightforward as those described for the laboratory studies. While the laboratory experiments provided as constant a set of exposure conditions as possible, the data required for defining comparable exposure conditions in the field were difficult, if not impossible, to collect. Consequently, the exposure-residue relationships established in the field were, by necessity, more qualitative than quantitative.

141. Although qualitative, estimates of BRH exposure in the field were necessary to establish exposure conditions for the laboratory-to-field comparison (i.e., establish when exposure conditions were similar in the laboratory and the field). The following generalizations are evident from the estimated BRH exposure concentrations: (a) the two independent methods, tissue residue and whole water analysis, provided remarkably similar estimates of BRH exposure and (b) there was a distinct exposure signal at 1 m above the bottom during and immediately postdisposal and that signal was transient, decreasing spatially and temporally postdisposal.

142. Comparison of the tissue residue and water chemistry estimates of BRH concentrations in CLIS indicated very good correspondence between the two. Several examples demonstrate this point. Tissue residue data (PCBs) from the T + 2 collection (Table 15) indicated that the BRH concentration was estimated to range between 1.4 and 0.8 mg/ℓ at the CNTR station. Water samples from the same station estimated the BRH concentration to range between 1.1 and 0.7 mg/ℓ (Table 16) using PCB values, and 1.3 and 0.7 using copper values. Approximately 1 month later, BRH concentration estimates, based on PCB tissue residues, were between 0.7 and 0.2 mg/ℓ at the CNTR station (T + 8, Table 15). The corresponding water chemistry estimates of BRH concentrations at the CNTR station ranged from 0.2 to 0.1 mg/ℓ, using PCBs, and 0.6 to 0.3 mg/ℓ, using copper. The similarity of these estimates indicates that exposure based on whole water chemistry concentrations and tissue residue concentrations tracked reasonably well.

143. The good relationship between the residue and water chemistry BRH estimates further supports the second generalization mentioned above: BRH exposure 1 m above the bottom was maximal immediately postdisposal and decreased over time. Spatially, both the PCB tissue residue and water

chemistry data indicated that BRH exposure decreased moving away from the CNTR station immediately postdisposal (Tables 15 and 16). This pattern persisted until T + 12, when tissue residues were similar at each station. The loss of the spatial differences in BRH exposures would suggest that exposure from the disposal mound was minimal after this collection period. Temporally, the maximum estimated concentration of BRH material 1 m above the bottom ranged between 1.4 and 0.8 mg/ℓ (tissue residues at T + 2, Table 15). At the time of the next collection this value decreased by approximately one-half, and continued to decrease over time.

144. While a range was calculated to estimate BRH exposure concentrations, it is interesting to note the temporal pattern of the high and low estimates. The high estimate of BRH exposure never reached zero, even in the later collections (i.e., T + 55, T + 116). This may indicate that background PCB levels in CLIS contributed to the BRH estimates, including those immediately postdisposal. The low estimate, calculated by subtracting the concentration at the REFS station, was assumed to remove the background concentration present in CLIS. Therefore, the low estimate, while providing a measure of relative difference between the stations, also may have provided the best estimate of actual BRH concentration. However, even using the high estimates, the data suggest that the integrated exposure of BRH material to *M. edulis*, 1 m above the bottom, was minimal at all the FVP stations and decreased rapidly following completion of the disposal operation.

145. The relationship between residue and effect in the field was not as direct as that in the laboratory. Several apparently contradictory results occurred between the residue data and the SFG data in the field. First, during the T + 2 collection, when maximum residues (i.e., exposures) occurred, no decrease in SFG was noted. Secondly, mussels retrieved from the CNTR station at T + 8, when residue concentrations were lower than at T + 2, did appear to exhibit an adverse SFG effect. These results suggested several alternative conclusions: (a) the highest field BRH exposures had no adverse effect on SFG in mussels, (b) an effect may have been observed if samples were not lost, (c) an "overshoot" occurred in the physiological response of the T + 2 mussels, or (d) temperature differences between collections may have caused differential sensitivity to BRH material.

146. First, it is possible that the BRH exposure concentration at T + 2 may have been insufficient to cause a reduction in SFG. The estimated maximum

BRH exposure at a station where mussels were deployed was between 1.4 and 0.8 mg/ℓ. If the lower estimate (0.8) for exposure was more accurate, it would represent about half that of the lowest suspended sediment concentration in the laboratory (1.5 mg/ℓ). Therefore, the signal in the field may have been "weaker" than that present in the laboratory experiments. This field level may have been below a possible threshold concentration required to elicit a physiological response, as suggested by the laboratory experiments.

147. Another possibility concerns the fact that the T + 8 collection was the first in which mussels were recovered from all four stations. Comparison of mussel SFG values from the REF station at T + 2, lost prior to biological sampling, with those from the 400E station would have presumably represented the widest range of residue concentrations from the field. It is possible that some biological effect may have been observed between these two stations; that, however, is only speculation. Future studies will include greater redundancy with respect to mussel deployments in order to minimize sample losses.

148. There is the possibility that a physiological "overshoot" occurred in the T + 2 mussels as a result of their removal from CLIS to clean water in the laboratory. Widdows, Donkin, and Evans (1985) reported that the SFG of mussels removed from chronic oil exposures returned to levels greater than control mussels. This reported physiological "overshoot" was greater in mussels exposed to higher concentrations of oil than to lower concentrations. While this phenomenon would account for the elevated SFG of the mussels at 400E, the time frame in the present study makes this unlikely. Widdows, Donkin, and Evans (1985) indicated that this overshoot occurred between 10 and 20 days after removal from the oil exposures. In the present study, SFG measurements were initiated on field mussels within 24 hr of collection from the field. Therefore, total recovery from field exposure to BRH in this short time period would be unlikely. Previous experiments involving field exposure of *M. edulis* to sewage sludge indicated that reduced SFG values persisted in mussels 7 days after they were returned to clean laboratory seawater.*

149. The most plausible explanation for the SFG results observed in mussels collected at T + 2 and T + 8 concerns possible differential effects due to temperature. Bayne (1976) reported that the SFG of *M. edulis* remains

* Unpublished data, William Nelson, SAIC, Narragansett, R. I.

relatively independent of temperature from 10° to 20° C. Above this range, physiological mechanisms responsible for metabolic compensation begin to break down. It is possible that the mussels collected at T + 8, when water temperatures were approximately 20° C, were less able to compensate for these conditions and thus exhibited an increased sensitivity to BRH material. Mussels at T + 2, when water temperature was 15° C, indicated no adverse SFG effect. These mussels may have been able to compensate physiologically for the slightly higher BRH concentrations.

150. A second point is that the clearance rates in the mussels from the CNTR station at T + 8 were reduced only slightly compared to mussels at the other three stations. In the laboratory, mussels exposed to 1.5 mg/ℓ for 28 days exhibited dramatic reductions in clearance rates compared to mussels exposed only to REF material. This estimated concentration of BRH in the field (0.7-0.2 mg/ℓ) may have been near the threshold level required to affect the mussels.

151. To summarize these two collections, mussels retrieved at T + 2 were unaffected by the estimated concentration of BRH suspended sediment present during this time period because this concentration may have been insufficient to cause a negative effect. Mussels collected at T + 8, while exposed to lower BRH concentrations, were slightly affected by the BRH material, as evidenced by lower clearance rates and SFG. Elevated temperatures (20° C) during this collection most likely increased the sensitivity of the mussels to BRH material.

152. After the T + 8 collection, tissue residue levels of PCB, PAH, and ethylan decreased with time for the subsequent monthly collections. In addition, SFG values were similar at all four stations during these collections. It would appear that these reduced residues did not adversely affect SFG even with the increased water temperatures during these collections (approximately 20° C). These data are consistent with the suggestion that most field concentrations of BRH suspended sediment were below that required to elicit a physiological response, and that the BRH concentrations present at the CNTR station at T + 8 (0.7-0.2 mg/ℓ) may have been very close to the suggested threshold concentration.

153. In addition to 1-month field deployments, other mussels were deployed for longer periods of time. Interpretation of these physiological data was more difficult than for the 1-month deployments. It followed that if it

was difficult to adequately explain exposures during the 1-month deployments, exposures of 3 months (T + 27, T + 43, T + 74) or 7 months (T + 21, T + 55) would increase the probability that even more unanticipated, unaccountable events might occur. For example, mussels collected at T + 21 were deployed in CLIS for 7 months. Mussels from REF exhibited a higher clearance rate than mussels from the other three stations. In addition, SFG values at the CNTR and 400E stations were lower than the REF station, while not different from the 1000E station. A comparison of the physiological results with the tissue residues indicated slightly elevated PCB and PAH levels in the mussels collected from the CNTR, 400E, and 1000E stations compared with the REF mussels.

154. These data may indicate exposure to BRH material during this deployment period as one possible explanation for the SFG results. In order for this to be true, however, some "resuspension event" would have had to occur at the CLIS disposal site. The negative physiological response was not present in the T + 15 mussels, collected 6 weeks earlier, and also absent in the T + 27 mussels, which were placed in the field at T + 15 and collected on T + 27. The cause of the observed response would have had to occur between T + 15 and T + 21, but disappear after that because it was missing at T + 27.

155. Another example of an unexplained SFG effect was the T + 43 collection. Mussels collected at the CNTR station exhibited a dramatic reduction in clearance rate, absorption efficiency, and SFG compared with the other two stations (the 1000E station was missing). These data indicate that the mussels at this station were impacted by something. Resuspension of bottom sediments may once again provide a possible explanation. This deployment occurred during the winter months when storms are typically stronger than during the summer and fall. If bottom sediments containing BRH material were resuspended and filtered out of the water by the mussels 1 m above the bottom, the clearance rate and SFG results could be explained based on the results of laboratory experiments. However, corresponding tissue residue concentrations were not elevated in the mussels from any station. In addition, not even laboratory exposures to 10 mg/ℓ of BRH suspended sediment affected absorption efficiencies in this manner. The exact cause of the observed effects cannot be explained based on BRH material alone; however, the magnitude of the response would suggest that it is not a measurement artifact.

156. The final collection (T + 116), a 1-month deployment, indicated that SFG values were not different among stations; however, they were all very

low. Several plausible reasons are presented. First, temperatures in CLIS were about 22° C, which may have stressed the animals. Secondly, there was an abnormal occurrence at the Narragansett Bay reference site at the time mussels were collected for deployment in CLIS. An incredible bloom of a small (<2-μ) algal species occurred throughout Narragansett Bay. This alga, present in concentrations of greater than 1 billion cells/litre, caused the mussels and some other bivalves in the Bay to cease feeding, resulting in mass mortalities in mussel populations. This condition was noted 1 month prior to the T + 116 collection (June 1985) and persisted until the middle of August 1985. As a result, mussels deployed at this time were not in the best physiological condition.

157. To summarize the field experiments, exposure data generated independently from water chemistry samples and tissue residues indicated that maximum exposure to BRH material occurred during the disposal operation and decreased rapidly thereafter. Of the range estimated for BRH concentrations in the field, the lower estimate may be closer to the actual value. The maximum estimated exposure in the field, between 1.4 and 0.8 mg/ℓ, was lower than the lowest exposure concentration in the laboratory (1.5 mg/ℓ). Elevated temperatures at T + 8 may have increased the sensitivity of mussels to BRH material, which would explain the reduced SFG of mussels at the CNTR station during this collection. Reduced residues after T + 8 were probably too low to elicit a negative physiological impact on the mussels. Therefore, the effect of BRH material on the SFG of mussels 1 m off the bottom was minimal. Mussels deployed in CLIS longer than 1 month indicated that there were apparent SFG differences on only two other occasions, T + 21 and T + 43, and both were difficult to attribute to the BRH material alone. From the field data, a BRH exposure concentration slightly greater than 1.4 to 0.8 mg/ℓ would be estimated to cause an adverse effect on SFG in mussels. This level may decrease as temperatures increase and mussels are less able to compensate for the BRH material.

Laboratory-to-Field Comparison

158. The purpose of the laboratory-to-field comparison was to expose mussels to BRH material in the laboratory and the field and compare the concentrations that produced a SFG effect in the mussels. The comparison of

laboratory and field data indicated one obvious fact: laboratory and field BRH exposures were different. The two independent estimates of BRH exposure indicated that the maximum exposure in the field was lower than BRH exposures in the laboratory. Cluster analysis of laboratory and field mussel residue data yielded results similar to those obtained in estimating the field exposures. That is, the mussel residues in the field were most similar to mussels exposed to reference sediment in the laboratory. Considering these data, discussion of the laboratory-field comparison will focus initially on when conditions (exposures and residues) were similar in the laboratory and field, and secondly on estimated concentrations of BRH suspended material required to affect SFG.

159. The exposure and residue data indicated that the most legitimate comparison between laboratory and field SFG data was between all field samples (with the exception of 400E at T + 2) and laboratory mussels exposed to 0-percent BRH. Furthermore, because temperature and season during the T = 0, T + 2, and T + 55 collections were most similar to the laboratory exposures, these SFG values should provide the most accurate laboratory-to-field comparison. The mean SFG value of mussels exposed to 0-percent BRH in the laboratory for 28 days was 7.2 J/hr. The field-exposed mussels exhibited SFG values of 9.5, 8.9, and 6.5 J/hr, respectively, for the T = 0 (1000E), T = 0 (REF), and T + 2 (1000E) collections. The similarity of these SFG values indicated that the relative physiological conditions of these mussels were the same when environmental conditions were most alike. The mussel collection at T + 55 also occurred in the spring at water temperatures similar to the laboratory exposures; however, these mussels had been deployed in CLIS for 8 months. The SFG values of these mussels, 3.0, 3.9, and 5.5 J/hr, were slightly lower than in the 1-month laboratory exposure (7.7 J/hr). This may be attributable to the difference in length of exposure.

160. The purpose of this qualitative comparison between laboratory and field was to establish whether similar exposures produced similar results. In light of the differences between the actual laboratory exposures (constant exposure levels, food supply, etc.) and field exposures (fluctuating particulate levels, food quantity, etc.), the SFG values between the two exposures were relatively similar.

161. A second aspect of the laboratory-to-field comparison was a qualitative evaluation of the estimate of BRH material required to produce a SFG

effect in the laboratory and field. The SFG, clearance rate, and actual growth measurements during laboratory experiments provided a clear signal that exposure to as little as 1.5 mg/ℓ of BRH material negatively affected *M. edulis*. From these data, an estimated BRH exposure approximating 1.5 mg/ℓ should be sufficient to adversely affect mussels exposed in the field. While the estimated maximum exposure concentration in the field (1.4-0.8 mg/ℓ, T + 2) did not appear to affect the mussels, an estimated lower concentration (0.7-0.2 mg/ℓ, CNTR station at T + 8) apparently did. This apparent contradiction was interpreted in terms of increased water temperatures during the T + 8 collection and consequently greater sensitivity of the mussels to BRH material. Because of the probable interactive effect of temperature, a qualitative estimate of the field BRH exposure concentration necessary to produce a SFG effect would be between 0 and 2.0 mg/ℓ.

162. Comparison of these two values indicated that the field estimate of 0.7 to 0.2 mg/ℓ was in the range predicted from laboratory experiments (less than or equal to 1.5 mg/ℓ). The possible existence of an effective threshold BRH concentration between 0 and 1.5 mg/ℓ, suggested by the laboratory dose-response curve, may help to explain the lack of effect in the field and the presence of one in the laboratory at comparable temperatures. In addition, the possibility of temperature-related sensitivity to this material would help to explain the presence of an effect at lower exposure levels. Nonetheless, the laboratory and field estimates provide a good qualitative comparison of the effects of BRH material on mussels.

PART V: CONCLUSIONS

163. The research described in this report evaluated the effects of a dredged material on the physiological condition of *M. edulis* after laboratory and field exposures. The results are as follows:

<u>a</u>. Laboratory dosing systems were successfully developed to expose mussels to relatively constant concentrations of suspended sediment.

<u>b</u>. A very good relationship was found between BRH laboratory exposure level and tissue residue concentration for higher molecular weight organic compounds such as PCBs. Lower molecular weight PAHs, such as phenanthrene, were apparently metabolized and/or excreted by the mussels.

<u>c</u>. An inverse relationship was observed in the laboratory between SFG and BRH exposure concentration. Likewise, SFG was inversely related to the tissue residue concentrations of some of the contaminants present in the dredged material.

<u>d</u>. Lower SFG values in BRH-exposed mussels were attributable to reduced clearance rates observed in the laboratory. In addition, mussels with lower SFG values exhibited reduced shell growth rates.

<u>e</u>. The results of the laboratory study indicated that the mussel residue concentrations were indicative of exposure conditions and that the SFG index was useful for measuring the subsequent biological effects of those exposures.

<u>f</u>. Independent estimates of field BRH concentrations, made from tissue residues as well as from water chemistry data, indicated that maximum exposure to mussels occurred during the disposal operation and decreased rapidly (2 weeks) thereafter. The maximum estimated concentration in the field (1.4 to 0.8 mg/ℓ) was lower than that in the lowest concentration used in the laboratory experiments (1.5 mg/ℓ).

<u>g</u>. The effect of the BRH exposures on the SFG of mussels, 1 m above the bottom, was minimal. Reductions in SFG, attributable to BRH exposure, occurred only one time, 8 weeks postdisposal. This effect was possibly due to increased sensitivity of mussels to dredged material as a result of elevated water temperatures.

<u>h</u>. The results of the field portion of this study indicated that the SFG of mussels was not affected because the BRH exposure concentrations were minimal.

<u>i</u>. A qualitative comparison between the SFG of mussels after laboratory and field exposures indicated that the estimated BRH concentration which affected SFG in the field (0.2 mg/ℓ) was similar to the range predicted from the laboratory experiments (1.5 mg/ℓ).

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APPENDIX A: CHEMICAL FORMULAS AND FIELD MUSSEL RESIDUE CONCENTRATIONS

Table A1

Chemical Contaminants Selected for Measurement in Both Field and Laboratory Studies

Chlorinated hydrocarbon pesticides

Polychlorinated biphenyls
Ethylan

Aromatic hydrocarbons ≥ molecular weight 166

Compound Class	Molecular Weight
Fluorene	166
C-1* Fluorene	180
C-2* Fluorene	194
C-3* Fluorene	208
C-4* Fluorene	222
Phenanthrene	178
Anthracene	178
C-1*Phenanthrene/anthracene	192
C-2*Phenanthrene/anthracene	206
C-3*Phenanthrene/anthracene	220
C-4*Phenanthrene/anthracene	234
Fluoranthene	202
Pyrene	202
C-1*Fluoranthene/pyrene	216
C-2*Fluoranthene/pyrene	230
C-3*Fluoranthene/pyrene	244
C-4*Fluoranthene/pyrene	258
Benzanthracene/chrysene**	228
C-1*Benzanthracene/chrysene**	242
C-2*Benzanthracene/chrysene**	256
C-3*Benzanthracene/chrysene**	270
C-4*Benzanthracene/chrysene**	284
Benzofluoranthenes	252
Benzo(e)pyrene	252
Benzo(a)pyrene	252
Perylene	252

(Continued)

* C-1, C-2, C-3, and C-4 refer to the number of methyl groups substituted somewhere in the parent compound.

** These names are representative of the class of polynuclear aromatic hydrocarbons (PAHs) measured at each molecular weight.

Table A1 (Concluded)

	Compound Class	Molecular Weight
	C-1*Benzopyrene/perylene**	266
	C-2*Benzopyrene/perylene**	280
	C-3*Benzopyrene/perylene**	294
	C-4*Benzopyrene/perylene**	308
	Benzoperylene**	276
	Dibenzanthracene**	278
	Coronene	300
	Dibenzocrysene**	302
Hetrocyclic aromatic compounds		
	Dibenzothiophene	184
	C-1*dibenzothiophene	198
	C-2*dibenzothiophene	212
	C-3*dibenzothiophene	226
	C-4*dibenzothiophene	240
Metals		
	Cadmium	
	Copper	
	Chromium	
	Iron	
	Lead	
	Manganese	
	Nickel	
	Zinc	

* C-1, C-2, C-3, and C-4 refer to the number of methyl groups substituted somewhere in the parent compound.

** These names are representative of the class of polynuclear aromatic hydrocarbons (PAHs) measured at each molecular weight.

Table A2

Complete Formulae for Calculating all SUM and CENT Variables

PSUM	=	POS166 + POS178 + POS202 + POS228 + POS252 + POS276 + POS278 + POS300 + POS302
HSUM	=	H1C166 + H2C166 + H3C166 + H4C166 + H1C178 + H2C178 + H3C178 + H4C178 + H1C202 + H2C202 + H3C202 + H4C202 + H1C228 + H2C228 + H3C228 + H4C228 + H1C252 + H2C252 + H3C252 + H4C252
SUM	=	POS166 + H1C166 + H2C166 + H3C166 + H4C166 + POS178 + H1C178 + H2C178 + H3C178 + H4C178 + POS202 + H1C202 + H2C202 + H3C202 + H4C202 + POS228 + H1C228 + H2C228 + H3C228 + H4C228 + POS252 + H1C252 + H2C252 + H3C252 + H4C252 + POS276 + POS278 + POS300 + POS302
PCENT	=	[POS166*166 + POS178*178 + POS202*202 + POS228*228 + POS252*252 + POS276*276 + POS278*278 + POS300*300 + POS302*302]/PSUM
HCENT	=	[H1C166*180 + H2C166*194 + H3C166*208 + H4C166*222 + H1C178*192 + H2C178*206 + H3C178*220 + H4C178*234 + H1C202*216 + H2C202*230 + H3C202*244 + H4C202*258 + H1C228*242 + H2C228*256 + H3C228*270 + H4C228*284 + H1C252*266 + H2C252*280 + H3C252*294 + H4C252*308]/HSUM
CENT	=	[POS166*166 + H1C166*180 + H2C166*194 + H3C166*208 + H4C166*222 + POS178*178 + H1C178*192 + H2C178*206 + H3C178*220 + H4C178*234 + POS202*202 + H1C202*216 + H2C202*230 + H3C202*244 + H4C202*258 + POS228*228 + H1C228*242 + H2C228*256 + H3C228*270 + H4C228*284 + POS252*252 + H1C252*266 + H2C252*280 + H3C252*294 + H4C252*308 + POS276*276 + POS278*278 + POS300*300 + POS302*302]/SUM

The sum of alkyl homologs of PAH molecular weight 178 (HOS178) is calculated according to the following formula:

HOS178 = H1C178 + H2C178 + H3C178 + H4C178

where

HOS178 = sum of C-1 to C-4 alkyl-substituted 178 PAHs

This statistic was chosen to describe the alkyl homologs because the 178 alkyl homologs are the most intense homologs within the Black Rock Harbor (BRH) PAH distribution and because they afford the greatest BRH to REFS concentration ratio. Alkyl homologs were included because of potential differences between them and parent PAHs.

Table A3

Tissue Residue Concentrations in Mussels from the T - 4 Field Collection in CLIS (22 Apr 83)*

Chemical Compound	Station CNTR	400E	1000E	REFS
Phenanthrene	210.0	117.0	98.0	38.0
Sum of 178 alkyl homologs	580.0	310.0	310.0	290.0
Fluoranthene	161.0	102.0	90.0	82.0
Benzo(a)pyrene	37.0	20.0	34.0	25.0
Ethylan	5.0	3.0	5.0	10.0
PCB as A1254	380.0	270.0	400.0	440.0
SUM of PAHs	2,600.0	1,520.0	1,650.0	1,380.0
CENTROID of PAHs	218.0	219.0	225.0	228.0
Copper	13.5	15.1	14.5	12.5
Cadmium	1.9	1.8	1.8	1.8
Chromium	1.8	3.8	2.2	1.6
Iron	370.0	1,400.0	530.0	340.0

* Units are nanograms per gram dry weight for the organic compounds and the statistic SUM, micrograms per gram dry weight for the inorganic elements, and molecular weight for the statistic CENTROID.

Table A4

Tissue Residue Concentrations in Mussels from the T + 0 Field Collection in CLIS (24 May 83).* The CNTR Station Was Not Deployed Because of the Dumping Operation, and the 400E Station Was Lost

	Station	
Chemical Compound	1000E	REFS
Phenanthrene	43.0	16.0
Sum of 178 alkyl homologs	1,440.0	290.0
Fluoranthene	161.0	52.0
Benzo(a)pyrene	100.0	18.0
Ethylan	102.0	9.0
PCB as A1254	1,080.0	500.0
SUM of PAHs	5,400.0	1,290.0
CENTROID of PAHs	230.0	232.0
Copper	16.5	10.9
Cadmium	2.0	2.3
Chromium	2.6	1.5
Iron	420.0	330.0

* Units are nanograms per gram dry weight for the organic compounds and the statistic SUM, micrograms per gram dry weight for the inorganic elements, and molecular weight for the statistic CENTROID.

Table A5

Tissue Residue Concentrations in Mussels from the T + 2 Field Collection in CLIS (07 June 83).* The CNTR Station Was Not Deployed Because of the Disposal Operation

Chemical Compound	Station 400E	Station 1000E	Station REFS
Phenanthrene	69.0	41.0	13.0
Sum of 178 alkyl homologs	1,900.0	970.0	540.0
Fluoranthene	290.0	126.0	72.0
Benzo(a)pyrene	210.0	118.0	51.0
Ethylan	71.0	39.0	17.0
PCB as A1254	1,440.0	1,020.0	630.0
SUM of PAHs	8,700.0	4,700.0	2,500.0
CENTROID of PAHs	232.0	234.0	233.0
Copper	16.9	15.6	10.8
Cadmium	2.3	2.3	1.9
Chromium	3.0	3.0	2.0
Iron	51[illegible]	560.0	560.0

* Units are nanograms per gram dry weight for the organic cor [illegible]ounds and the statistic SUM, micrograms per gram dry weight for the inorganic elements, and molecu[illegible]ar weight for the statistic CENTROID.

Table A6

Tissue Residue Concentrations in Mussels from the T + 8 Field Collection in CLIS (10 Jul 83)*

Chemical Compound	Station CNTR	400E	1000E	REFS
Phenanthrene	11.0	14.0	9.0	7.0
Sum of 178 alkyl homologs	350.0	340.0	193.0	105.0
Fluoranthene	45.0	46.0	31.0	23.0
Benzo(a)pyrene	40.0	50.0	18.0	20.0
Ethylan	22.0	20.0	7.0	1.0
PCB as A1254	700.0	740.0	620.0	480.0
SUM of PAHs	1,870.0	2,100.0	1,020.0	760.0
CENTROID of PAHs	234.0	236.0	231.0	240.0
Copper	10.1	9.6	11.5	4.4
Cadmium	1.9	2.0	1.3	0.9
Chromium	1.4	1.4	3.2	0.8
Iron	340.0	370.0	820.0	240.0

* Units are nanograms per gram dry weight for the organic compounds and the statistic SUM, micrograms per gram dry weight for the inorganic elements, and molecular weight for the statistic CENTROID.

Table A7

Tissue Residue Concentrations in Mussels from the T + 12 Field Collection in CLIS (10 Aug 83)*

Chemical Compound	Station			
	CNTR	400E	1000E	REFS
Phenanthrene	17.0	10.0	9.0	8.0
Sum of 178 alkyl homologs	250.0	160.0	96.0	65.0
Fluoranthene	41.0	28.0	20.0	15.0
Benzo(a)pyrene	41.0	17.0	16.0	13.0
Ethylan	9.0	8.0	3.0	1.0
PCB as A1254	640.0	660.0	550.0	570.0
SUM of PAHs	1,600.0	940.0	710.0	530.0
CENTROID of PAHs	237.0	236.0	239.0	240.0
Copper	5.3	5.6	7.5	5.8
Cadmium	0.9	0.9	1.2	1.1
Chromium	1.0	0.7	1.6	0.7
Iron	164.0	167.0	450.0	177.0

* Units are nanograms per gram dry weight for the organic compounds and the statistic SUM, micrograms per gram dry weight for the inorganic elements, and molecular weight for the statistic CENTROID.

Table A8

Tissue Residue Concentrations in Mussels from the T + 15 Field Collection in CLIS (06 Sep 83)*

Chemical Compound	Station CNTR	400E	1000E	REFS
Phenanthrene	13.0	9.0	10.0	6.0
Sum of 178 alkyl homologs	370.0	230.0	210.0	43.0
Fluoranthene	57.0	38.0	33.0	14.0
Benzo(a)pyrene	53.0	45.0	28.0	7.0
Ethylan	10.0	6.0	4.0	1.0
PCB as A1254	870.0	630.0	640.0	550.0
SUM of PAHs	2,100.0	1,540.0	1,240.0	350.0
CENTROID of PAHs	236.0	239.0	237.0	238.0
Copper	7.7	6.0	8.0	5.8
Cadmium	1.0	1.1	1.1	0.9
Chromium	1.2	0.9	1.1	0.9
Iron	260.0	179.0	290.0	260.0

* Units are nanograms per gram dry weight for the organic compounds and the statistic SUM, micrograms per gram dry weight for the inorganic elements, and molecular weight for the statistic CENTROID.

Table A9

Tissue Residue Concentrations in Mussels from the T + 21 Field Collection in CLIS (18 Oct 83)*

Chemical Compound	Station			
	CNTR	400E	1000E	REFS
Phenanthrene	12.0	11.0	11.0	10.0
Sum of 178 alkyl homologs	132.0	101.0	88.0	46.0
Fluoranthene	33.0	25.0	22.0	16.0
Benzo(a)pyrene	24.0	9.0	17.0	9.0
Ethylan	2.0	2.0	1.0	0.0
PCB as A1254	540.0	680.0	570.0	420.0
SUM of PAHs	1,000.0	670.0	700.0	400.0
CENTROID of PAHs	240.0	234.0	239.0	238.0
Copper	22.1	16.3	15.1	16.4
Cadmium	5.1	4.4	4.8	5.0
Chromium	2.3	2.6	2.2	2.2
Iron	440.0	540.0	420.0	480.0

* Units are nanograms per gram dry weight for the organic compounds and the statistic SUM, micrograms per gram dry weight for the inorganic elements, and molecular weight for the statistic CENTROID.

Table A10

Tissue Residue Concentrations in Mussels from the T + 27 Field Collection in CLIS (29 Nov 83).* The CNTR Station Was Missing at the Time of Collection

Chemical Compound	Station 400E	1000E	REFS
Phenanthrene	18.0	10.0	8.0
Sum of 178 alkyl homologs	230.0	117.0	86.0
Fluoranthene	68.0	36.0	37.0
Benzo(a)pyrene	39.0	32.0	19.0
Ethylan	3.0	1.0	0.0
PCB as A1254	540.0	380.0	450.0
SUM of PAHs	1,820.0	1,150.0	860.0
CENTROID of PAHs	240.0	244.0	240.0
Copper	16.4	21.0	23.3
Cadmium	3.2	3.4	3.6
Chromium	2.5	3.6	3.3
Iron	570.0	920.0	920.0

* Units are nanograms per gram dry weight for the organic compounds and the statistic SUM, micrograms per gram dry weight for the inorganic elements, and molecular weight for the statistic CENTROID.

Table A11

Tissue Residue Concentrations in Mussels from the T + 43 Field Collection in CLIS (20 Mar 84).* Metals Were Not Measured for These Samples. The 1000E Station Was Missing at the Time of Collection

Chemical Compound	Station CNTR	400E	REFS
Phenanthrene	16	18	17
Sum of 178 alkyl homologs	94	78	70
Fluoranthene	28	26	24
Benzo(a)pyrene	8	4	7
Ethylan	2	1	1
PCB as A1254	350	330	280
SUM of PAHs	510	460	450
CENTROID of PAHs	229	230	231

* Units are nanograms per gram dry weight for the organic compounds and the statistic SUM, and molecular weight for the statistic CENTROID.

Table A12

Tissue Residue Concentrations in Mussels from the T + 55 Field Collection in CLIS (05 June 84).* Metals Were Not Measured in These Samples. The CNTR Station Was Missing at the Time of Collection

	Station		
Chemical Compound	400E	1000E	REFS
Phenanthrene	6	6	4
Sum of 178 alkyl homologs	89	91	54
Fluoranthene	25	31	18
Benzo(a)pyrene	6	7	6
Ethylan	0	1	0
PCB as A1254	540	490	550
SUM of PAHs	520	550	370
CENTROID of PAHs	234	235	236

* Units are nanograms per gram dry weight for the organic compounds and the statistic SUM, and molecular weight for the statistic CENTROID.

Table A13

Tissue Residue Concentrations in Mussels from the T + 116 Field Collection in CLIS (13 Aug 85)*

Chemical Compound	Station CNTR	400E	1000E	REFS
Phenanthrene	3.0	6.0	3.0	3.0
Sum of 178 alkyl homologs	79.0	124.0	80.0	58.0
Fluoranthene	18.0	27.0	24.0	19.0
Benzo(a)pyrene	18.0	40.0	22.0	17.0
Ethylan	1.0	2.0	1.0	0.0
PCB as A1254	310.0	350.0	450.0	440.0
SUM of PAHs	700.0	1,270.0	810.0	620.0
CENTROID of PAHs	242.0	243.0	241.0	244.0
Copper	8.5	7.5	6.9	7.6
Cadmium	1.5	1.3	1.3	1.3
Chromium	1.2	1.1	0.9	0.9
Iron	290.0	260.0	220.0	220.0

* Units are nanograms per gram dry weight for the organic compounds and the statistic SUM, micrograms per gram dry weight for the inorganic elements, and molecular weight for the statistic CENTROID.

APPENDIX B: COMPARISON OF SELECTED CONTAMINANTS IN BRH AND REF SEDIMENTS

The contaminant-specific analysis of the BRH and REF sediments is presented in summary form for the representative subset of chemical compounds discussed in this report. These analyses demonstrate clearly the differences in contaminant concentration between the two sediments.

Table B1

Concentrations of the Ten Selected Contaminants and Two Summary Statistics for Both BRH and REF Sediments. Means ± Standard Deviations

Chemical Compound	Sediment* BRH	Sediment* REF
Phenanthrene	5,200 ± 1,820 (8)**	85 ± 16 (12)
Sum of 178 alkyl homologs	29,000 ± 8,400 (8)	100 ± 19 (12)
Fluoranthene	6,500 ± 1,400 (8)	240 ± 31 (12)
Benzo(a)pyrene	4,000 ± 950 (8)	251 ± 27 (12)
Ethylan	3,900 ± 750 (8)	0 ± -- (12)
PCB as A1254	7,000 ± 1,560 (8)	39 ± 4 (12)
SUM of PAHs	146,000 ± 31,000 (8)	4,500 ± 490 (12)
CENTROID of PAHs	232.7 ± 1.6 (8)	249.2 ± 1.6 (12)
Copper	2,900 ± 310 (54)	60 ± 3 (45)
Cadmium	24 ± 1.0 (54)	0.23 ± 0.04 (45)
Chromium	1,480 ± 104 (54)	50 ± 15 (45)
Iron	31,000 ± 2,800 (54)	21,000 ± 1,400 (45)

* Units are nanograms per gram dry weight for the organic compounds and the statistic SUM, micrograms per gram dry weight for the inorganic elements, and molecular weight for the statistic CENTROID.

** (N) = number of replicates.