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SOLID PHASE BIOASSAY AND
BIOACCUMULATION OF SEDIMENT
PRESENT AT THE ALCATRAZ
DISPOSAL SITE

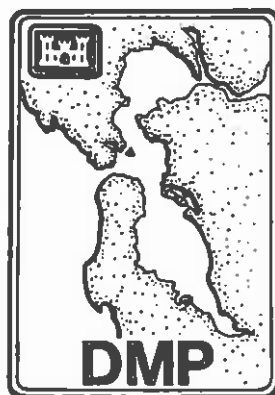
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PREFACE

The San Francisco District of the Corps of Engineers has recently begun a program to review and evaluate existing dredging and disposal operations in San Francisco Bay. The program, called the Disposal Management Program (DMP) will address the following issues:

1. Material accumulation at the Alcatraz disposal site (SF-11).
2. Periodic bathymetric monitoring of the two north bay disposal sites: Carquinez Straits (SF-9), and San Pablo Bay (SF-10).
3. Availability and feasibility of potential alternate disposal options.
4. Fate of dredged material as a result of open-water disposal.
5. Ocean disposal site designation.

Completion of the program is scheduled for July 1988.

The following report, "Solid Phase Bioassay and Bioaccumulation of Sediment Present at the Alcatraz Disposal Site" documents the results of toxicity testing of sediments present at the surface of the Alcatraz Disposal Site during the summer of 1985.

for more information on the study or the DMP in general, contact Mr. Les Tong at (415) 974-0439 or ((FTS) 454-0439, or Ms. Jody Zaitlin at (415) 974-0442 or (FTS) 454-0442, Environmental Branch, San Francisco District Corps of Engineers, 211 Main St., San Francisco, California 94105-1905.

SUMMARY

A series of solid-phase bioassays and bioaccumulation analyses was conducted to assess the toxicity of sediment from the Alcatraz Disposal Site in San Francisco Bay. Survival of amphipods (Rhepoxynius abronius) exposed for 10 days to reference sediment (collected near the south tower of the Golden Gate Bridge) was significantly higher than survival of amphipods exposed to sediment from the Alcatraz Disposal Site. Survival of speckled sanddab (Citharichthys stigmaeus) and the clam (Macoma nasuta) exposed to reference sediment and sediment from the Alcatraz Disposal Site for 20 days was not significantly different.

Tissues of clams that survived exposure to sediment from the Alcatraz Site during the 20-day bioassay did not contain significantly elevated concentrations of any of the 18 measured xenobiotic constituents (arsenic, cadmium, chromium, copper, lead, mercury, selenium, silver, zinc, aldrin, dieldrin, chlordane and related compounds, DDT and derivatives, endrin, hexachlorocyclohexane, PCBs, toxaphene, and petroleum hydrocarbons) as compared to tissues of reference organisms.

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1. INTRODUCTION

The major objective of this evaluation is to assess the toxicity of sediment present at the Alcatraz Disposal Site. The evaluation consists of five principal sections in addition to the Introduction. The first section, which precedes the Introduction, summarizes the results of this study. The second section reviews the methods and materials employed in the evaluation. The third section presents important results of the evaluation. The fourth section discusses results of the evaluation and the fifth section contains references cited in the report.

The evaluation contains four appendices. Appendix A details laboratory procedures employed for preparing sediment and conducting bioassays. The appendix also serves as a quality-control document. Appendix B contains all raw bioassay data. Only data directly relevant to the ecological evaluation of the proposed discharge operation were presented in the main body of the report. Appendix C presents quality control and quality assurance information, and Appendix D presents results of grain-size distribution analyses performed on reference and control sediment.

2. METHODS AND MATERIALS

Sediment from the Alcatraz Disposal Site was collected from five randomly selected stations in the site, which is circular in shape and 2,000 ft in diameter. The site is located south of Alcatraz Island (coordinates for the center of the circle are: 37°49'17"N, and 122°25'23"W). Sampling station locations (Figure 1) were determined by drawing north-south and east-west axes through the center of the site, numbering each axis from 1-1,000, and selecting pairs of random numbers (one on each axis) between 0 and 1,000. Station 1 was located approximately 750 ft north of the center of the site, Station 2 was located near the north-west perimeter, Station 3 was situated near the center of the site, Station 4 was located approximately 500 ft east of the center of the site, and Station 5 was positioned approximately 700 ft south of the center of the site.

Reference sediment was collected from a potential disposal site located to the east of the south tower of the Golden Gate Bridge (coordinates of the center of the site are: 37°48'55"N and 122°28'13"W). This site (which has been designated the "South Tower Site") is identical in size and shape to the Alcatraz Disposal Site. This reference location was selected because the South Tower Site is located near the Alcatraz Disposal Site in an uncontaminated area of similar grain-size distribution (Kinnetic Laboratories, Inc., 1985), and is not located downstream from the predominant currents at the disposal site. Samples were collected at five randomly selected stations (Figure 1). The first three stations were clustered in an area approximately 400-600 ft north to north-west of the center of the site. Station 4 was situated slightly to the west of the site's center and the fifth station was located to the east of the center, near the perimeter of the site.

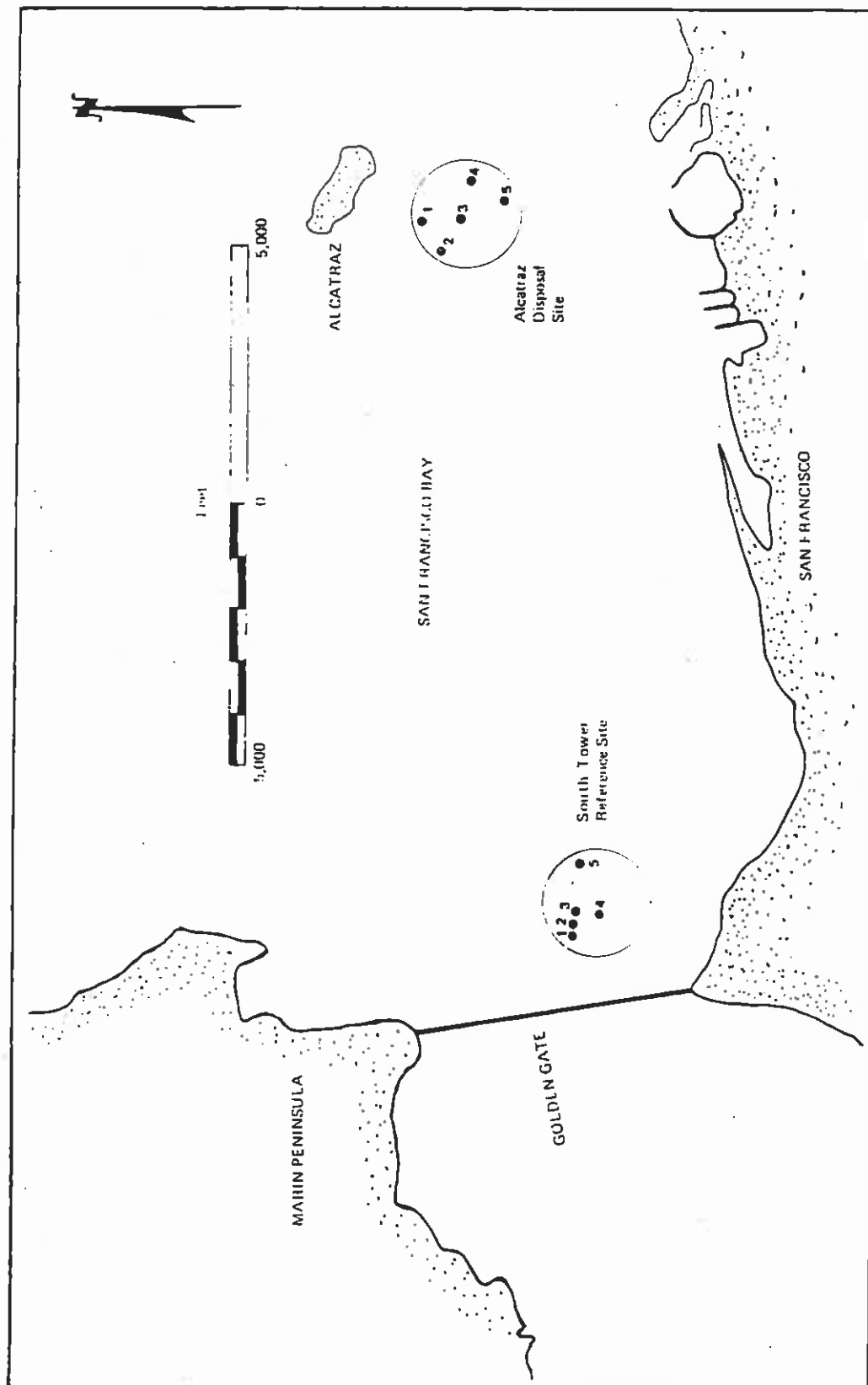


Figure 1. Station locations for collection of sediment from the Alcatraz Disposal Site and the South Tower Reference Site in San Francisco Bay.

Sediment from the Alcatraz Disposal Site and reference sediment were collected during 09:55-11:10 and 07:30-09:45 respectively, on October 15, 1985. Material was collected by Timothy J. Ward and Robert L. Boeri of ENSECO, from the vessel F/V LEONILDA. At the Alcatraz Site, bottom temperature was 17.5°C and salinity was 30.5 ppt at the time of sample collection. At each sample station 1-5 grab samples were collected to a depth of 1 ft using a stainless steel Van Veen sampler. Samples were placed in completely filled polyethylene buckets. Buckets were placed in ice-filled coolers and delivered by overnight courier to ENSECO's Aquatic Toxicology Laboratory in Cambridge, Massachusetts, where they were placed in cold storage at 2-4°C. Prior to use in tests, Alcatraz Disposal Site sediment and reference sediment were well mixed.

Sediment was prepared for biological testing according to procedures described in Appendix B of the U.S. EPA and COE manual for dredged material (U.S. EPA and U.S. Army COE, 1977). Two separate solid phase bioassays were performed with sediment from the Alcatraz Disposal Site. The first study was performed with the amphipod (Rhepoxynius abronius), and the second study was performed with the speckled sanddab (Citharichthys stigmaeus), and the clam (Macoma nasuta).

The solid phase bioassay with the amphipod was performed according to methods authored by Swartz et al (1985). Amphipods were obtained from a commercial supplier in Newport, Oregon, who collected them from the field. After arrival at ENSECO's laboratory, animals were placed in aquaria containing control sediment (material gathered from the organism collection location in Oregon) and artificial seawater, where they were acclimated to test conditions for 3 days (temperature = 17.5°C and salinity = 30.5 ppt). Amphipods were tested in 1-liter glass beakers containing a 2-cm deep (approximately 240 g) layer of sediment and artificial seawater. Control beakers contained

a 2-cm layer of control sediment, reference beakers contained a 2-cm layer of material from the South Tower Reference Site and treatment beakers contained a 2 cm layer of material from the Alcatraz Disposal Site. The test continued for 10 days under static conditions during which the number of amphipods appearing on the surface was noted daily, as was the temperature, pH and dissolved oxygen concentration of each treatment. At the end of the test the number of survivors and the number of amphipods unable to burrow in control sediment after 1 hr was determined.

The solid phase bioassays with speckled sanddab and clams were conducted according to guidelines presented in Appendix F of the EPA and COE manual for dredged material (U.S. EPA and U.S. Army COE, 1977). Speckled sanddab (mean standard length, 6 cm) were obtained from a commercial supplier in Dillon Beach, California and clams (mean length, 3.5 cm) were obtained from a commercial supplier in Newport, Oregon. Fish and clams were acclimated to test conditions for 4-10 days prior to initiation of the bioassays. Both species were tested in the same aquaria. Each treatment (disposal site, reference and control) was replicated 10 times with 10 organisms per replicate. The disposal site material treatment consisted of a 15-mm layer of sediment from the Alcatraz Site on a 30-mm layer of reference sediment from the South Tower Site. The reference aquaria contained a 15-mm layer of control sediment (collected from the subtidal zone off Manchester, Massachusetts by Jane L. Freedman, ENSECO, on October 28, 1985) above a 30-mm layer of reference sediment, and the control aquaria contained a 45-mm layer of control sediment. Testing temperature was $17.5 \pm 2.0^{\circ}\text{C}$, and salinity of the artificial seawater was adjusted to 30.5 ppt. Water exchange was by the flow-through method at the rate of six tank volumes per 24-hr. The tests were conducted for 20 days, during which time temperature of the environmental chamber in which tests were performed was continuously recorded, and pH, and dissolved oxygen concentration were measured daily in each

aquarium. Each aquarium was supplied with approximately 25 ml frozen plankton or frozen San Francisco Bay brine shrimp daily throughout the 20-day test.

At the conclusion of the solid phase bioassays with speckled sanddab and clams, all surviving clams from each aquarium were placed in an aquarium containing clean, sediment-free water and allowed to void their digestive systems for a period of 2 days. During this time, fecal material was removed from aquaria. At the end of the 2-day period, all samples of organisms from replicates 1 and 2, 3 and 4, 5 and 6, 7 and 8, and 9 and 10 were pooled and split into approximately equal amounts. One of these subsamples was placed in a polyethylene clean bag and frozen for later analyses for metals. The second subsample was put in solvent-rinsed aluminum foil and frozen for analyses for organics. Prior to being chemically analyzed, clams were thawed and shells were removed with acid-rinsed plastic utensils (metal analyses) or solvent-rinsed metal utensils (organic analyses).

Tissue samples were analyzed for nine metals -- arsenic (As), cadmium (Cd), chromium (Cr), copper (Cu), lead (Pb), mercury (Hg), selenium (Se), silver (Ag), and zinc (Zn) -- according to procedures described by Goldberg (1976) and the EPA (1979). In the analyses for As, Cd, Cr, Cu, Pb, Se, Ag, and Zn, an aliquot of wet, homogenized tissue (approximately 5 g) was placed in a 100-ml tall-form Pyrex beaker with 5 ml of concentrated, Instra-analyzed (J.T. Baker Co.) nitric acid and refluxed without boiling until the tissue was completely digested (6-24 hr). Following digestion, the sample was evaporated to dryness. Then, additional nitric acid (1-2 ml) and 30% Ultrex (J.T. Baker Co.) hydrogen peroxide (1-2 ml) were added to the beaker, and the sample was heated until oxidative frothing subsided. At this time, the sample was cooled, diluted to volume with deionized, distilled water, and analyzed by

graphite-furnace atomic absorption spectrophotometry (AAS). Procedural blanks and standards were evaluated using the same methods employed for tissue samples. For the analyses for Hg, a separate aliquot of wet, homogenized tissue (about 5 g) was placed in a 300-ml glass BOD bottle. Approximately 15-20 ml of concentrated, Instra-analyzed sulfuric acid was placed in the bottle and the sample was heated at 55°C in a water bath until the tissue was completely digested (2 hr). After cooling of the sample, 100 ml of deionized, distilled water and 1-2 g of Instra-analyzed potassium permanganate were added to the bottle. The resulting solution was analyzed by cold-vapor AAS after addition of reducing agents (10% hydroxylamine hydrochloride and 10% stannous sulfate). Procedural blanks and standards were assessed by the same methods used for tissue samples.

Tissue samples were analyzed for aldrin, dieldrin, chlordane and related compounds, DDT and derivatives, endrin, hexachlorocyclohexane, PCBs, and toxaphene according to procedures described by the Food and Drug Administration (1968), Sections 211.13, 211.14. The fat was extracted using petroleum ether and a tissue homogenizer. The extracts were combined, concentrated, and placed into a 125 ml separator. A total of 30 ml CH_3CN saturated with petroleum ether was added and the mixture shaken vigorously for 1 min. The layers were allowed to separate and the CH_3CN was drained into a 1-liter separator containing 650 ml of water, 40 ml of saturated NaCl solution and 100 ml of petroleum ether. The petroleum ether solution was extracted in a 125 ml separator with three additional 30-ml portions of CH_3CN saturated with petroleum ether, shaken vigorously for 1 min each time, and then all extracts were combined in the 1-liter separator. The separator was then held horizontally and the solution was mixed thoroughly for 30-45 sec. The layers were allowed to separate and the aqueous layer was drained into a second 1-liter separator. Then 100 ml of petroleum ether was added to the second separator, shaken vigorously for 15 sec, and

allowed to separate. The aqueous layer was discarded, and the petroleum ether was combined with the petroleum ether in the original separator and washed with two, 100-ml portions of water. The washings were discarded and the layer of petroleum ether was drained through a column of anhydrous Na_2SO_4 , (25 mm x 50 mm long) into a 500 ml Kuderna-Danish concentrator. The separator and column were then rinsed with three 10-ml portions of petroleum ether. The combined extract and rinses were evaporated to 5-10 ml in the Kuderna-Danish concentrator for transfer to a Florisil column, a 22-mm i.d. column that contained 100 mm (after settling) activated Florisil topped with about 10 mm anhydrous Na_2SO_4 . The column was prewet with 40-50 ml of petroleum ether. The Kuderna-Danish concentrator was placed with a volumetric graduated collection vessel under the column to receive the eluate. The petroleum ether solution of the sample extract was then transferred through the column at about 5 ml/min. The container was rinsed with two, 5-ml portions of petroleum ether, and the rinsings were transferred to the column. The walls of the chromatographic tube were rinsed with petroleum ether. The column was eluted at about 5 ml/min with 200 ml of 6% ethyl ether/petroleum ether eluant. The receivers were changed and eluted at about 5 ml/min with 200 ml of 15% ethyl ether/petroleum ether eluant. Each eluate was concentrated to a final volume of 0.5 ml with a rotary evaporator and analyzed by packed-column gas chromatography and electron-capture detection, employing a Hewlett-Packard Model 5840A or 5880 instrument equipped with a Ni^{63} detector. The column, a 6-ft x 2-mm I.D. glass instrument packed with 5% SP2401 or 1.95% SP2401 and 1.5% SP2250, was held isothermally at 200°C. The peaks were quantified by comparing retention times and peak areas to those of external standards.

Tissue samples were analyzed for petroleum hydrocarbons according to procedures described by the U.S. EPA (1971), Crump-Wiesner et al. (1974), the U.S. Food and Drug

Administration (1977), and Warner (1976). Tissue samples (5-20 g wet wt.) were placed in 50-ml centrifuge tubes, to which 10-ml aliquots of 10 N potassium hydroxide and high-purity methanol, and 5 μ g of an internal standard (androstane) were added. After sealing with nitrogen gas, the tubes were placed in a water bath at 80°C for 4 hr (tubes were shaken every 30 min). This saponification process digested the tissue, thereby releasing petroleum hydrocarbons. Three 20-ml portions of high-purity hexane were used to extract the original compounds of interest from the methanol/potassium hydroxide digestate. The water fraction was then discarded. The three extracts were combined, dried over a small volume (10 g) of sodium sulfate, and concentrated to 1 ml by flash evaporation. The extracts were then fractionated using column chromatography (1 g sodium sulfate, 6.5 g of 7.5% deactivated alumina, and 1 g sodium sulfate) as follows. The 1-ml concentrate was charged to the top of the column and the column was eluted with 25 ml of hexane. The hexane was concentrated to 2 ml by flash evaporation, and further concentrated to 0.5 ml under a stream of purified nitrogen. An aliquot of the column-chromatographic fraction was analyzed for petroleum hydrocarbons by glass capillary gas chromatography and flame ionization detection, employing a Hewlett-Packard Model 5840A instrument. The column, a 0.25-mm I.D. x 30-m SE30 glass capillary fused silica column (J&W Scientific), was temperature-programmed from 60°C to 275°C at 10°/min. The areas of the resolved and unresolved components were measured by electronic integration and planimetry, respectively, and compared to the areas of an internal standard (androstane) to determine the concentration of petroleum hydrocarbons.

Results of the bioassay and bioaccumulation tests were interpreted by statistical techniques recommended by the U.S. EPA and U.S. Army COE (1977). When warranted, each data set generated in the studies was evaluated by Cochran's procedure to

determine if variances of the data were homogeneous. Because all variances from analyzed data sets were homogeneous, a parametric, unpaired "t" test was used to determine if a significant difference existed between reference organisms and organisms exposed to sediment from the Alcatraz Disposal Site. In all statistical tests, the symbols "*" and "ns" are used to denote significant and nonsignificant differences (at the 0.05 probability level), respectively.

3. RESULTS

Material from the Alcatraz Disposal Site was heterogeneous, consisting of fine black silt at some locations and sand with broken shells at other locations (visual determination). Material from the South Tower Reference Site and control sediment (both from Oregon and Massachusetts) consisted of fine sand with some silt. Results of grain-size distribution analyses of reference sediment and Massachusetts control sediment appear in Appendix D and results of chemical analysis of Massachusetts control sediment appears in Appendix C.

3.1 Solid Phase Bioassay with the Amphipod (*Rhepoxynius abronius*)

Data produced by the solid phase bioassay with amphipods are summarized in Table 1 and presented graphically in Figure 2. Mean survival of organisms exposed for 10 days to sediment from the Alcatraz Disposal Site was equal to 80.0%, as compared to survival of animals exposed to control and reference sediment, which were 93.0% and 92.0%, respectively. All raw data from this test appears in Appendix B (Tables B1 and B2). Statistical analyses of survival data for amphipods exposed for 10 days to reference sediment and sediment from the Alcatraz Site (Table 2) indicates that survival of organisms exposed to Alcatraz Disposal Site sediment was significantly less (0.05 probability level) than survival of organisms exposed to reference sediment.

Throughout the 10-day exposure, fewer amphipods were visible on the surface of sediment from the Alcatraz Disposal Site than either reference or control sediment. Statistical analysis of emergence data (total number of animals present on the surface

Table 1. Survival and sublethal effects of amphipods (Rhepoxynius abronius) exposed for 10 days to control sediment, reference sediment, and Alcatraz Disposal Site sediment

Treatment	Replicate	Total number of amphipods emerged from sediment (day 0-10)	Number of survivors after 10-day exposure	Number of survivors unable to rebury
Control sediment	1	7	19	0
	2	3	18	0
	3	2	18	0
	4	2	19	0
	5	1	19	0
	Mean ($\bar{x}$)	3.0	18.6	0.0
	Std. Dev.	2.3	0.5	0.0
Reference sediment	1	8	20	0
	2	1	17	0
	3	0	20	0
	4	7	17	0
	5	1	18	0
	Mean ($\bar{x}$)	3.4	18.4	0.0
	Std. Dev.	3.8	1.5	0.0
Alcatraz Disposal Site sediment	1	3	14	1
	2	3	18	1
	3	2	19	0
	4	3	15	2
	5	1	14	1
	Mean ($\bar{x}$)	2.4	16.0	1.0
	Std. Dev.	0.9	2.3	0.7

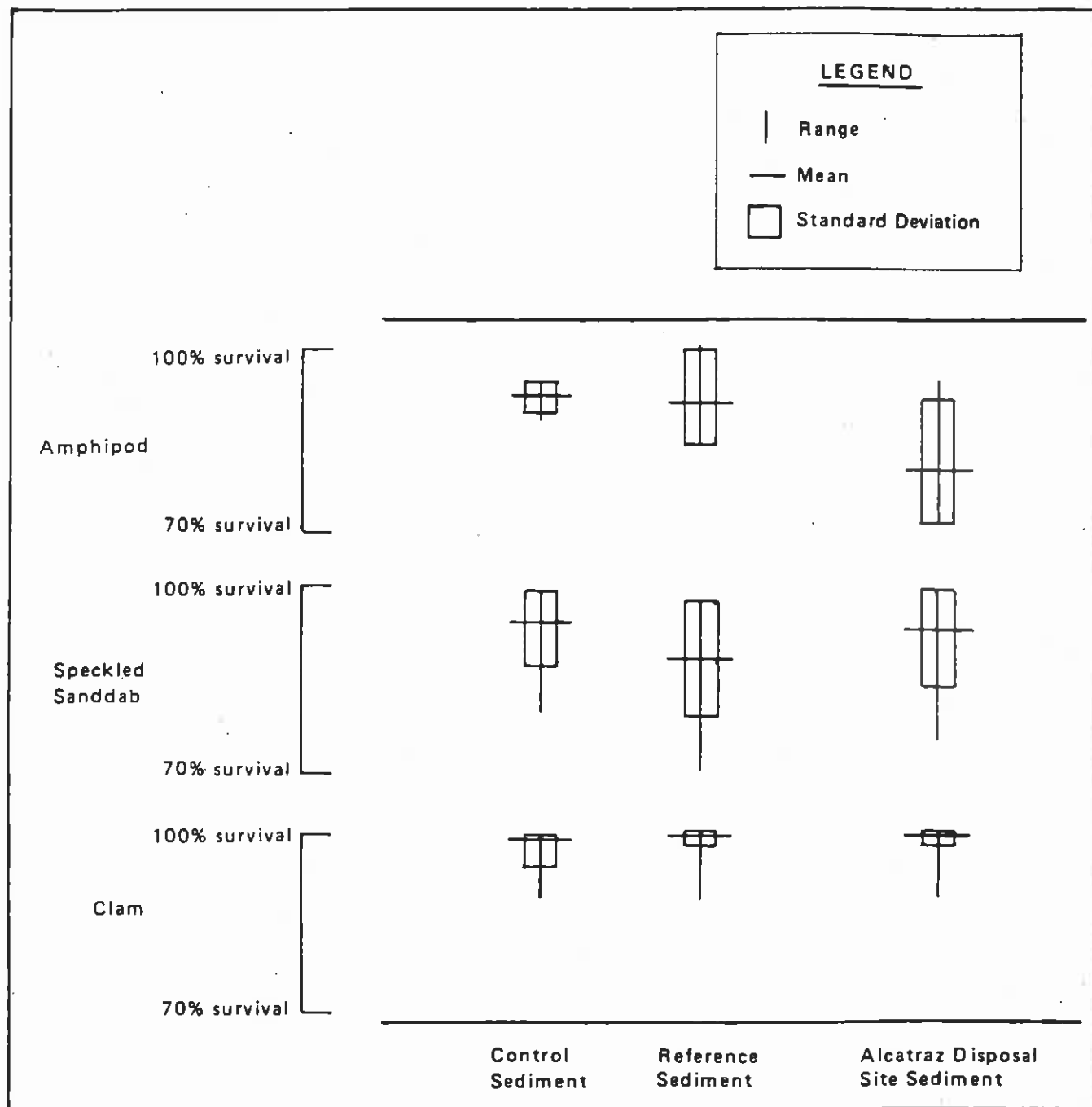


Figure 2. Results of solid phase bioassays with the amphipod (*Rhepoxynius abronius*), speckled sanddab (*Citharichthys stigmaeus*) and the clam (*Macoma nasuta*).

Table 2. Statistical analyses of survival and emergence of amphipods (Rhepoxynius abronius), and survival of speckled sanddab (Citharichthys stigmaceus), and the clam (Macoma nasuta) exposed to sediment from the Alcatraz Disposal Site and reference sediment

Organism	Cochran's test for homogeneity of variances of survival data				Parametric unpaired "t" test (reference sediment vs Alcatraz Disposal Site sediment	
	Treatment	Mean ($\bar{x}$)	Variance (S^2)	$C_{(cal)}^a$	$t_{(cal)}^b$	
Amphipod (survival)	Alcatraz site	16.0	5.5	0.71 ns	1.92 *	
	Reference site	18.4	2.3			
Amphipod (emergence)	Alcatraz site	2.4	Further analysis not warranted --			
	Reference site	3.4	($\bar{x}$ for Alcatraz site is less than $\bar{x}$ for reference sediment)			
Speckled sanddab (survival)	Alcatraz site	9.3	Further analysis not warranted --			
	Reference site	9.0	($\bar{x}$ for Alcatraz site is greater than $\bar{x}$ for reference sediment)			
Clam (survival)	Alcatraz site	9.9	Further analysis not warranted --			
	Reference site	9.9	($\bar{x}$ for Alcatraz site is equal to $\bar{x}$ for reference sediment)			

^aThe value for $C_{(cal)}$ is compared to $C_{(tab)}$, which equals 0.91 for 0.05 probability level, $k = 2$ and $v = 4$. Variances are considered to be homogeneous (ns) if $C_{(cal)} \leq C_{(tab)}$. Otherwise, variances are considered to be heterogeneous (*).

^bThe value for $t_{(cal)}$ is compared to $t_{(tab)}$, which equals 1.86 for 0.05 probability level, one-tailed hypothesis, and $df = 8$. Mean survival of organisms is considered to be nonsignificantly different (ns) if $t_{(cal)} \leq t_{(tab)}$. Otherwise, the difference is considered to be significant (*).

during the 10-day test) for amphipods exposed to reference sediment and sediment from the Alcatraz Site (Table 2) is not warranted since fewer amphipods emerged from Alcatraz Disposal Site sediment than from reference sediment. Five of the 80 surviving amphipods from the Alcatraz Disposal Site sediment exposure were unable to rebury in clean sediment at the end of the 10-day exposure, as opposed to survivors from the control and reference exposure which all reburied.

3.2 Solid Phase Bioassays with Speckled Sanddab (*Citharichthys stigmaeus*) and the clam (*Macoma nasuta*)

Data produced by solid phase bioassays with speckled sanddab and clams are summarized in Table 3 and Figure 2. Mean survival of speckled sanddab ranged from 90% in reference sediment to 95% in control sediment. Survival of clams was greater than 98% in all three treatments (control, reference and disposal site). Statistical analysis of survival data (Table 2) is unwarranted for both the speckled sanddab and clam, because mean survival of organisms exposed to sediment from the Alcatraz Disposal Site was greater than survival of organisms exposed to reference sediment. It can be concluded therefore that based on the results of these two bioassays, sediment from the Alcatraz Disposal Site is not significantly more toxic than the naturally occurring sediment for the species tested.

3.3 Bioaccumulation Studies

Concentrations of arsenic, cadmium, chromium, copper, lead, mercury, selenium, silver, zinc, aldrin, dieldrin, chlordane and related compounds, DDT and derivatives, endrin, hexachloro-cyclohexane, PCBs, toxaphene, and petroleum hydrocarbons in

Table 3. Survival of speckled sanddab (*Citharichthys stigmaeus*) and the clam (*Macoma nasuta*) exposed for 20 days to control sediment, reference sediment, and Alcatraz Disposal Site sediment

Organism	Repli- cate	Number of survivors		
		Control Sediment	Reference Sediment	Alcatraz Disposal Site Sediment
Speckled sanddab	1	10	8	9
	2	10	10	10
	3	9	9	9
	4	10	9	9
	5	9	8	10
	6	10	9	10
	7	9	10	9
	8	10	9	10
	9	8	9	10
	10	10	9	7
	Mean ($\bar{x}$)	9.5	9.0	9.3
	Std. Dev.	0.7	0.7	0.9
Clam	1	10	10	10
	2	10	10	10
	3	10	9	10
	4	10	10	9
	5	10	10	10
	6	8	10	10
	7	10	10	10
	8	10	10	10
	9	10	10	10
	10	10	10	10
	Mean ($\bar{x}$)	9.8	9.9	9.9
	Std. Dev.	0.6	0.3	0.3

tissues of clams that survived the 20-day solid phase bioassay are presented in Table 4. Bioaccumulation analyses that yielded results above detection limits are presented in Figure 3. Statistical analysis of data sets in which tissue concentrations in organisms exposed to sediment from the Alcatraz Disposal Site are higher than concentrations in tissues of reference organisms appears in Table 5.

Concentrations of aldrin, dieldrin, endrin, hexachlorocyclohexane, PCBs and toxaphene were below detection levels in tissues of clams exposed to all three sediments (control, reference, and Alcatraz Disposal Site). Concentrations of the remaining tested chemicals -- arsenic, cadmium, chromium, copper, lead, mercury, selenium, silver, zinc, chlordane and related compounds, DDT and derivatives, and petroleum hydrocarbons were lower in tissues of organisms exposed to sediment from the Alcatraz Site than in reference organism tissue except in the case of two metals -- chromium and zinc. Statistical analysis of these two data sets indicates that no statistically significant differences exist between reference animals and those exposed to sediment from the Alcatraz Disposal Site.

Table 4. Concentrations of arsenic (As), cadmium (Cd), chromium (Cr), copper (Cu), lead (Pb), mercury (Hg), selenium (Se), silver (Ag), zinc (Zn), aldrin, dieldrin, chlordane and related compounds, DDT and derivatives, endrin, hexachlorocyclohexane, PCBs, toxaphene, and petroleum hydrocarbons in tissues of clams (*Macoma nasuta*) that survived 20-day exposure to the solid phase of control sediment, reference sediment, and sediment from the Alcatraz Disposal Site

Chemical	Repli- cate	Concentration of chemical in tissues (µg/g, wet wt.) ^a		
		Control sediment	Reference sediment	Alcatraz Disposal Site sediment
Arsenic	1	1.1	2.1	1.4
	2	1.4	2.1	0.89
	3	1.8	1.5	2.3
	4	1.3	1.4	1.2
	5	1.6	1.6	1.0
	Mean	1.4	1.7	1.4
	Std. Dev.	0.3	0.3	0.6
Cadmium	1	0.019	0.041	0.017
	2	0.040	0.10	0.012
	3	0.037	0.040	0.031
	4	0.058	0.025	0.028
	5	0.18	0.040	0.014
	Mean	0.067	0.049	0.020
	Std. Dev.	0.065	0.029	0.009
Chromium	1	<0.10	<0.10	<0.10
	2	<0.11	0.16	<0.097
	3	<0.10	<0.10	0.18
	4	<0.097	<0.098	<0.096
	5	<0.099	<0.098	<0.10
	Mean	<0.101	0.111	0.115
	Std. Dev.	0.005	0.027	0.037
Copper	1	1.6	2.9	1.1
	2	4.4	3.4	1.7
	3	1.8	3.6	2.7
	4	2.2	1.1	1.6
	5	1.6	2.6	1.6
	Mean	2.3	2.7	1.7
	Std. Dev.	1.2	1.0	0.6

Table 4. Continued

Chemical	Repli- cate	Concentration of chemical in tissues ($\mu\text{g/g}$, wet wt.) ^a		
		Control sediment	Reference sediment	Alcatraz Disposal Site sediment
Lead	1	0.23	0.32	0.20
	2	0.32	0.41	0.17
	3	0.21	0.28	0.44
	4	0.35	0.33	0.25
	5	0.19	0.20	0.27
	Mean	0.26	0.31	0.27
	Std. Dev.	0.07	0.08	0.10
Mercury	1	0.014	0.017	0.014
	2	0.012	0.017	0.014
	3	0.022	0.022	0.024
	4	0.016	0.012	0.019
	5	0.016	0.040	0.018
	Mean	0.016	0.022	0.018
	Std. Dev.	0.004	0.011	0.004
Selenium	1	0.13	0.16	<0.10
	2	0.18	0.21	<0.097
	3	0.17	0.15	0.19
	4	0.17	0.098	0.11
	5	<0.098	0.16	0.13
	Mean	0.150	0.156	0.125
	Std. Dev.	0.035	0.040	0.038
Silver	1	0.023	0.049	0.068
	2	0.091	0.056	0.042
	3	0.029	0.038	0.024
	4	0.020	0.017	0.028
	5	0.018	0.031	0.017
	Mean	0.036	0.038	0.036
	Std. Dev.	0.031	0.015	0.020
Zinc	1	9.9	20	15
	2	15	14	9.0
	3	20	16	23
	4	12	11	14
	5	15	12	21
	Mean	14.4	14.6	16.4
	Std. Dev.	3.8	3.6	5.6

Table 4. Continued

Chemical	Repli- cate	Concentration of chemical in tissues ($\mu\text{g/g}$, wet wt.) ^a		
		Control sediment	Reference sediment	Alcatraz Disposal Site sediment
Aldrin	1	<0.0001	<0.0001	<0.0001
	2	<0.0001	<0.0001	<0.0001
	3	<0.0001	<0.0001	<0.0001
	4	<0.0001	<0.0001	<0.0001
	5	<0.0001	<0.0001	<0.0001
	Mean	<0.0001	<0.0001	<0.0001
	Std. Dev.	0.0000	0.0000	0.0000
Dieldren	1	<0.0001	<0.0001	<0.0001
	2	<0.0001	<0.0001	<0.0001
	3	<0.0001	<0.0001	<0.0001
	4	<0.0001	<0.0001	<0.0001
	5	<0.0001	<0.0001	<0.0001
	Mean	<0.0001	<0.0001	<0.0001
	Std. Dev.	0.0000	0.0000	0.0000
Chlordane and related compounds	1	<0.0001	<0.0001	<0.0001
	2	<0.0001	<0.0001	<0.0001
	3	<0.0001	<0.0001	<0.0001
	4	<0.0001	0.0083	<0.0001
	5	<0.0001	0.0030	<0.0001
	Mean	<0.0001	0.0023	<0.0001
	Std. Dev.	0.0000	0.0036	0.0000
DDT and derivatives	1	<0.015	<0.015	<0.015
	2	<0.015	<0.015	<0.015
	3	<0.015	0.017	<0.015
	4	<0.015	<0.015	<0.015
	5	<0.015	<0.015	<0.015
	Mean	<0.015	0.015	<0.015
	Std. Dev.	0.000	0.009	0.000
Endrin	1	<0.0001	<0.0001	<0.0001
	2	<0.0001	<0.0001	<0.0001
	3	<0.0001	<0.0001	<0.0001
	4	<0.0001	<0.0001	<0.0001
	5	<0.0001	<0.0001	<0.0001
	Mean	<0.0001	<0.0001	<0.0001
	Std. Dev.	0.0000	0.0000	0.0000

Table 4. Continued

Chemical	Repli- cate	Concentration of chemical in tissues ($\mu\text{g/g}$, wet wt.) ^a		
		Control sediment	Reference sediment	Alcatraz Disposal Site sediment
Hexachlorocyclo- hexane	1	<0.0001	<0.0001	<0.0001
	2	<0.0001	<0.0001	<0.0001
	3	<0.0001	<0.0001	<0.0001
	4	<0.0001	<0.0001	<0.0001
	5	<0.0001	<0.0001	<0.0001
	Mean	<0.0001	<0.0001	<0.0001
	Std. Dev.	0.0000	0.0000	0.0000
PCBs	1	<0.002	<0.002	<0.002
	2	<0.002	<0.002	<0.002
	3	<0.002	<0.002	<0.002
	4	<0.002	<0.002	<0.002
	5	<0.002	<0.002	<0.002
	Mean	<0.002	<0.002	<0.002
	Std. Dev.	0.0000	0.0000	0.0000
Toxaphene	1	<0.0001	<0.0001	<0.0001
	2	<0.0001	<0.0001	<0.0001
	3	<0.0001	<0.0001	<0.0001
	4	<0.0001	<0.0001	<0.0001
	5	<0.0001	<0.0001	<0.0001
	Mean	<0.0001	<0.0001	<0.0001
	Std. Dev.	0.0000	0.0000	0.0000
Petroleum hydrocarbons	1	0.10	0.40	<0.10
	2	0.20	0.10	0.20
	3	0.30	0.30	<0.10
	4	0.50	<0.10	0.10
	5	<0.10	0.10	0.20
	Mean	0.24	0.20	0.14
	Std. Dev.	0.17	0.14	0.05

^aData sets in which mean tissue concentration in organisms exposed to sediment from the Alcatraz Disposal Site is higher than mean concentration in reference organisms are enclosed by boxes. Statistical analysis of the enclosed data sets appears in Table 5.

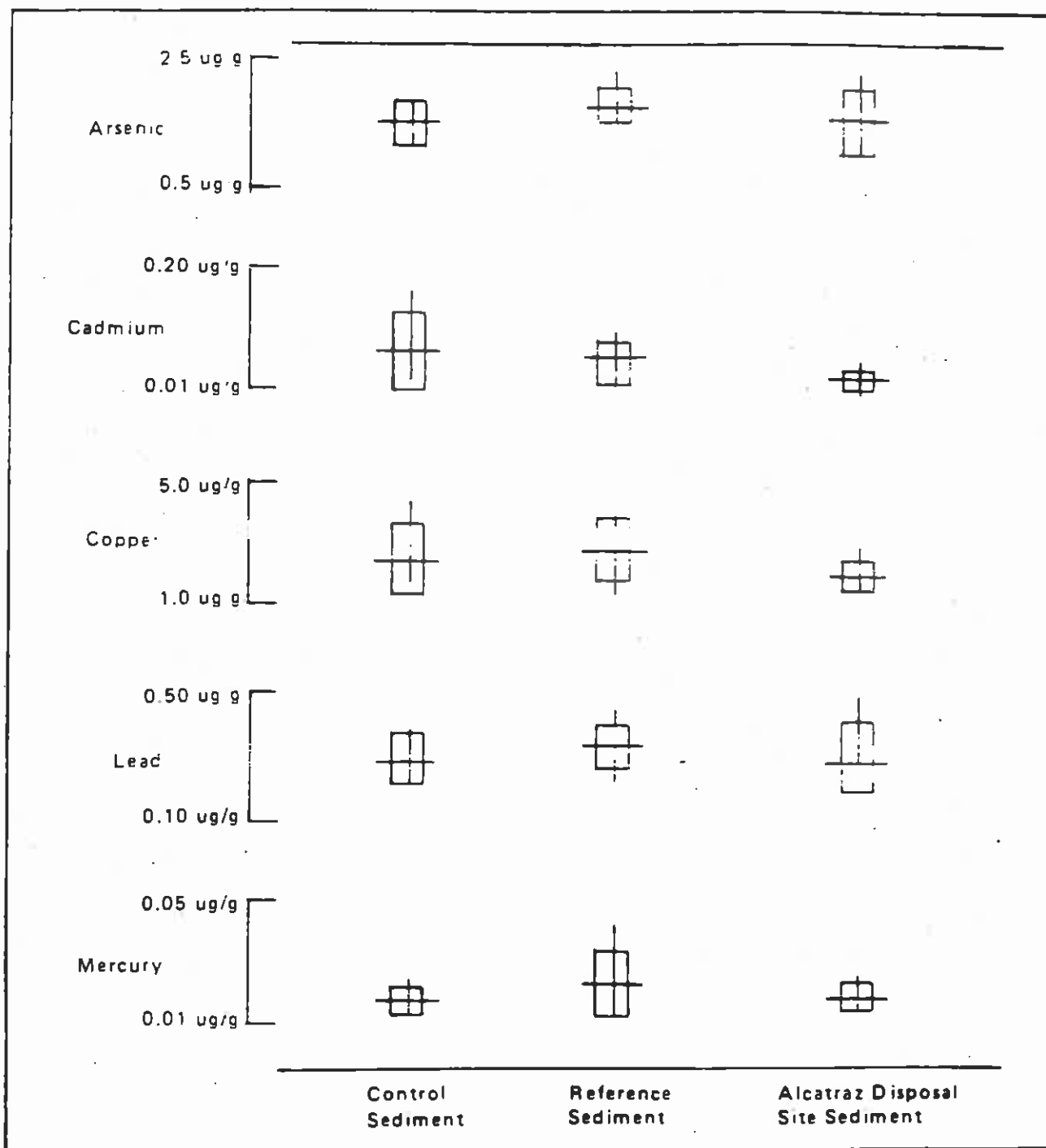


Figure 3. Concentration of arsenic (As), cadmium (Cd), copper (Cu), lead (Pb), mercury (Hg), silver (Ag), and zinc (Zn) in tissues of clams (*Macoma nasuta*) that survived 20-day exposure to control sediment, reference sediment, and sediment from the Alcatraz Disposal Site.

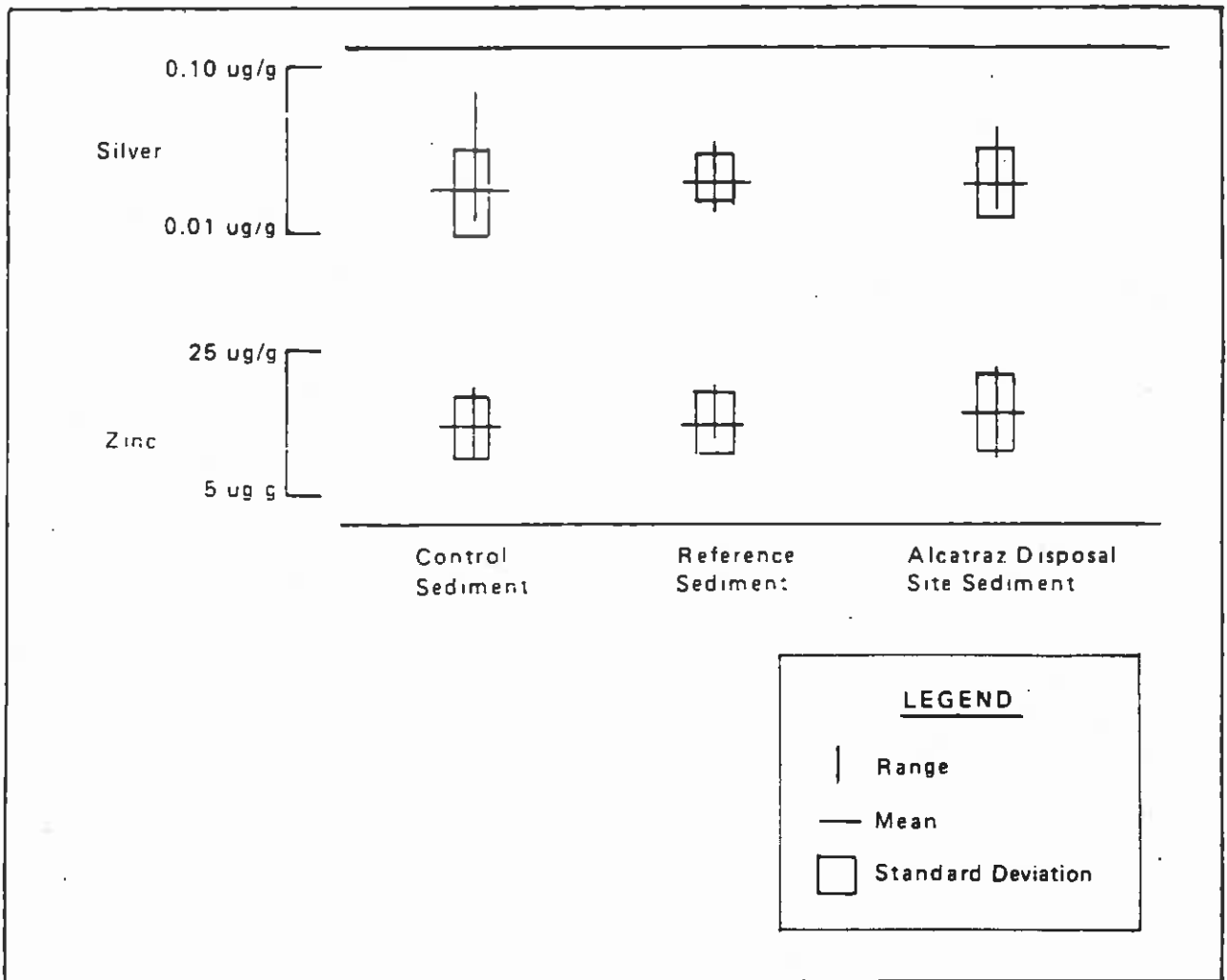


Figure 3 Continued.

Table 5. Statistical analyses of selected data in bioaccumulation study -- data sets in which mean tissue concentration in clams (*Macoma nasuta*) exposed to sediment from the Alcatraz Disposal Site is higher than mean concentration in reference organisms

Chemical constituent	Cochran's test for homogeneity of variances of chemical data				Parametric unpaired "t" test (reference sediment vs. Alcatraz Disposal sediment)
	Treatment	Mean ($\mu\text{g/g}$)	Variance	$C_{(\text{cal})}^a$	$t_{(\text{cal})}^b$
Chromium	Alcatraz Site	0.115	0.0013	0.65 ns	0.17 ns
	Reference Site	0.111	0.0007		
Zinc	Alcatraz Site	16.4	31.8	0.71 ns	0.60 ns
	Reference Site	14.6	12.8		

^aThe value for $C_{(\text{cal})}$ is compared to $C_{(\text{tab})}$, which equals 0.91 for 0.05 probability level, $k = 2$, and $v = 4$. Variances are considered to be homogeneous (ns) if $C_{(\text{cal})} \leq C_{(\text{tab})}$. Otherwise, variances are considered to be heterogeneous (*).

^bThe value for $t_{(\text{cal})}$ is compared to $t_{(\text{tab})}$, which equals 1.86 for 0.05 probability level, one-tailed hypothesis, and $df = 8$. Mean tissue concentrations are considered to be nonsignificantly different (ns) if $t_{(\text{cal})} \leq t_{(\text{tab})}$. Otherwise, the difference is considered to be significant (*).

4. DISCUSSION

If results of the solid phase bioassay performed with amphipods are judged according to criteria set forth by the EPA and COE (1977), the difference between survival of amphipods exposed to reference sediment and sediment from the Alcatraz Disposal Site would indicate that the material from the Alcatraz Site has a potential for causing environmentally unacceptable impacts on benthic organisms.¹ However, because this test procedure is not specifically recommended by the EPA and COE in their manual, interpretation of results according to the manual's criteria may be inappropriate. The authors of the amphipod bioassay method (Swartz et al, 1985) include no guidance for evaluating the toxicity of sediment based on results of this test. Mearns, et al (unpublished manuscript) did attempt to define toxic and nontoxic sediments through an interlaboratory study using this amphipod bioassay. They concluded that sediments which allow 76-87% survival after a 10-day exposure should be considered marginally toxic (a "gray area" between highly toxic and relatively nontoxic sediments). They suggest that these marginally toxic sediments, such as material from the Alcatraz Disposal Site, should be classified as toxic or nontoxic based on emergence of amphipods from the sediment during the 10-day exposure. Using this criterion, sediment from the Alcatraz Disposal Site would be classified as

¹Paragraph 37, page F17, Appendix F of the EPA and COE manual for dredged material (U.S. EPA and U.S. Army COE, 1977) states that a solid phase has "real potential for causing environmentally unacceptable impacts on benthic organisms if difference in mean survival between animals in the control and test sediments is statistically significant and greater than 10 percent."

nontoxic since greater emergence occurred in both the control and reference sediment exposures than in the exposure to sediment from the Alcatraz Site.

The bioassay studies performed with speckled sanddab and clams (tests performed according to EPA and COE [1977] procedures for evaluating the acceptability of oceanic disposal of dredged material) indicate that the material is not more toxic than naturally occurring sediments. In addition, all of the bioaccumulation tests performed during the assessment indicate no potential for xenobiotic constituents of the material to accumulate in the human food chain.²

Two major points concerning the assessment of potential bioaccumulation must be emphasized. The estimates could underestimate the likelihood of bioaccumulation since the 20-day exposure period employed in the evaluations may not be sufficiently long to allow chemical equilibrium to be established in the tissues of test organisms, i.e., organisms exposed to sediment may have been accumulating chemical constituents at the time that the bioaccumulations tests were terminated. Conversely, the assessments may overestimate the potential for bioaccumulation in that the 2-day depuration period employed in the tests may be too short to allow organisms to completely purge their digestive systems of chemical constituents.

²Paragraph 25, page G11, Appendix G of the EPA and COE manual for dredged material (U.S. EPA and U.S. Army COE, 1977) states that there is "no indication of potential bioaccumulation from [the solid phase of] the dredged material [if there are] no statistical differences between tissue concentration in the reference substrate controls and the dredged material."

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APPENDIX A

LABORATORY PROCEDURES FOR PREPARING SEDIMENT AND CONDUCTING BIOASSAYS

Procedure	Date of Implementation of Procedure	Certifications of Performance of Procedures		
		Aquatic Toxicologist	Aquatic Toxicologist	Laboratory Director
1. Store control sediment (CS), reference sediment (RS), and Alcatraz Disposal Site sediment (AS) at 2-4°C in separate containers. Mix sediment in each container as thoroughly as possible.	CS 10/21/85 and 10/28/85	<u>Jan. J. J.</u>	<u>Robert Bain</u>	<u>Patrick J. W.</u>
	AS 10/15/85	"	"	"
	RS 10/15/85	"	"	"

Solid Phase Bioassays with Amphipods

Bioassays should be initiated by October 29, 1985 (2 weeks after October 15, 1985, earliest date of sediment collection).

2. Remove CS, RS and AS from storage and wet sieve through 0.5-mm mesh. Place nonliving material remaining on sieve in appropriate containers.	10/24	"	"	"
3. Allow sediment to settle for 4 hr.	10/24	"	"	"
4. Decant water and resieve sediment through 0.5 mm mesh. Use ASW (30.5 ppt) for sieving. Allow sediment to settle for 24 hr, decant water, and mix sediment as thoroughly as possible.	10/24	"	"	"
5. Assign treatments (CS, RS, and AS) and replicates (6 per treatment) to 1-liter glass beakers.	10/25	"	"	"

Procedure	Date of Implementation of Procedure	Certifications of Performance of Procedures		
		Aquatic Toxicologist	Aquatic Toxicologist	Laboratory Director
6. Randomly position beakers in environmental chamber maintained at $17.5 \pm 2^{\circ}\text{C}$.	<u>10/25</u>	"	"	"
7. Add ASW to each beaker to the 750 ml mark.	<u>10/25</u>	"	"	"
8. Allow beakers to equilibrate for 24 hr.	<u>10/25-10/26</u>	"	"	"
9. Select amphipods from holding tanks containing ASW and control sediment and randomly distribute 20 into each of 15 culture dishes.	<u>10/26</u>	"	"	"
10. Randomly distribute contents of culture dishes into 5 of 6 beakers per treatment.	<u>10/26</u>	"	"	"
11. Increase water level in each beaker to the 950 ml mark.	<u>10/26</u>	"	"	"
12. Replace any amphipod that has not burrowed into sediment within 1 hr.	<u>10/26</u>	"	"	"
13. Perform the following activities:				
<u>Every day after introduction of amphipods into beakers</u>				
• Record temperature, dissolved oxygen, and pH in the beaker without amphipods for each treatment (CS, RS, and AS).	Day 0 <u>10/26</u>	"	"	"
	Day 1 <u>10/27</u>	"	"	"
	Day 2 <u>10/28</u>	"	"	"

Procedure	Date of Implementation of Procedure	Certifications of Performance of Procedures		
		Aquatic Toxicologist	Aquatic Toxicologist	Laboratory Director
● Record obvious mortality and the number of amphipods present on the sediment surface. (Do not remove dead amphipods.)	Day 3 <u>10/29</u>	"	"	"
	Day 4 <u>10/30</u>	"	"	"
	Day 5 <u>10/31</u>	"	"	"
	Day 6 <u>11/01</u>	"	"	"
	Day 7 <u>11/02</u>	"	"	"
	Day 8 <u>11/03</u>	"	"	"
	Day 9 <u>11/04</u>	"	"	"
	Day 10 <u>11/05</u>	"	"	"
14. At end of 10-day testing period, sieve sediment in each beaker and count live animals.	<u>11/05</u>	"	"	"
15. Place surviving amphipods in culture dish with 2-cm layer of control sediment and ASW. Note number unable to burrow in 1 hr.	<u>11/05</u>	"	"	"

Solid Phase Bioassays with Speckled Sanddab and Clams

Bioassays should be initiated by October 29, 1985 (2 weeks after October 15, 1985, earliest date of sediment collection).

16. Remove CS and RS from storage and wet sieve through 1-mm mesh. Use minimum volume of artificial seawater [ASW] of salinity 30.5 ppt for sieving purposes. Place nonliving material remaining on sieve in appropriate containers.

10/29

"

"

"

Procedure	Date of Implementation of Procedure	Certifications of Performance of Procedures		
		Aquatic Toxicologist	Aquatic Toxicologist	Laboratory Director
17. Mix CS and RS and allow to settle for 6 hr.	<u>10/29</u>	<u>"</u>	<u>"</u>	<u>"</u>
18. Decant ASW and mix CS and RS as thoroughly as possible.	<u>10/29</u>	<u>"</u>	<u>"</u>	<u>"</u>
19. Assign treatments (CS, RS and AS) and replicates (10 per treatment) to aquaria.	<u>10/29</u>	<u>"</u>	<u>"</u>	<u>"</u>
20. Randomly position aquaria in environmental chamber maintained at $17.5 \pm 2^{\circ}\text{C}$.	<u>10/29</u>	<u>"</u>	<u>"</u>	<u>"</u>
21. Partially fill aquaria with ASW.	<u>10/29</u>	<u>"</u>	<u>"</u>	<u>"</u>
22. Place 30 mm of CS in each control aquaria and 30 mm of RS in reference and treatment aquaria. Fill 1st aquarium to ~10 mm, then 2nd aquarium to ~10 mm, ..., and finally last aquarium to ~10 mm. Repeat sequence until aquaria are filled to ~20 mm. Repeat sequence again until aquaria are filled to ~30 mm. This procedure will help to ensure that CS and RS in all aquaria is homogeneous. Store remaining CS at $2-4^{\circ}\text{C}$ for later use.	<u>10/29</u>	<u>"</u>	<u>"</u>	<u>"</u>
23. Replace ASW 1 hr after CS and RS have been added to aquaria. Do not disturb sediment during replacement.	<u>10/29</u>	<u>"</u>	<u>"</u>	<u>"</u>

Procedure	Date of Implementation of Procedure	Certifications of Performance of Procedures		
		Aquatic Toxicologist	Aquatic Toxicologist	Laboratory Director
24. Select clams from holding tanks and randomly distribute 10 into each of 30 culture dishes.	<u>10/29</u>	<u>"</u>	<u>"</u>	<u>"</u>
25. Randomly distribute contents of culture dishes into aquaria.	<u>10/29</u>	<u>"</u>	<u>"</u>	<u>"</u>
26. If necessary, replace 75% of ASW 24 hr after animals are introduced into aquaria.	<u>Not necessary</u>	<u>"</u>	<u>"</u>	<u>"</u>
27. Acclimate animals for 48 hr. During this time period, remove dead animals and replace with live animals.	<u>10/29-10/31</u>	<u>"</u>	<u>"</u>	<u>"</u>
28. During acclimation period, remove appropriate volumes of AS from storage, composite samples, and wet-sieve sample through 1-mm mesh. Use minimum volume of ASW for sieving purposes. Place nonliving material remaining on sieve in container.	<u>10/31</u>	<u>"</u>	<u>"</u>	<u>"</u>
29. Mix AS and allow to settle for 6 hr.	<u>10/31</u>	<u>"</u>	<u>"</u>	<u>"</u>
30. Decant ASW and mix AS as thoroughly as possible.	<u>10/31</u>	<u>"</u>	<u>"</u>	<u>"</u>
31. Place 15 mm of AS in all but control and reference aquaria. Employ basic strategy identified in Step 22.	<u>10/31</u>	<u>"</u>	<u>"</u>	<u>"</u>

Procedure	Date of Implementation of Procedure	Certifications of Performance of Procedures		
		Aquatic Toxicologist	Aquatic Toxicologist	Laboratory Director
32. Remove remaining CS from storage. Warm to test temperature. Add 15 mm of CS to each control and reference aquaria. Employ basic strategy identified in Step 22.	<u>10/31</u>	<u>"</u>	<u>"</u>	<u>"</u>
33. Turn on flow-through seawater system 1 hr after addition of AS and final addition of CS.	<u>10/31</u>	<u>"</u>	<u>"</u>	<u>"</u>
34. Select speckled sanddab from holding tank and randomly distribute 10 into each culture dish.	<u>10/31</u>	<u>"</u>	<u>"</u>	<u>"</u>
35. Randomly distribute contents of culture dishes into aquaria.	<u>10/31</u>	<u>"</u>	<u>"</u>	<u>"</u>
36. Perform the following activities:				
<u>Every day after introduction of speckled sanddab into aquaria</u>				
● Record dissolved oxygen, and pH in each aquarium.	Day 0	<u>10/31</u>	<u>"</u>	<u>"</u>
	Day 1	<u>11/01</u>	<u>"</u>	<u>"</u>
	Day 2	<u>11/02</u>	<u>"</u>	<u>"</u>
● Record obvious mortality, and unusual behavior patterns of animals.	Day 3	<u>11/03</u>	<u>"</u>	<u>"</u>
	Day 4	<u>11/04</u>	<u>"</u>	<u>"</u>
	Day 5	<u>11/05</u>	<u>"</u>	<u>"</u>
	Day 6	<u>11/06</u>	<u>"</u>	<u>"</u>
● Feed frozen shrimp to organisms in each aquarium	Day 7	<u>11/07</u>	<u>"</u>	<u>"</u>
	Day 8	<u>11/08</u>	<u>"</u>	<u>"</u>

Procedure	Date of Implementation of Procedure	Certifications of Performance of Procedures		
		Aquatic Toxicologist	Aquatic Toxicologist	Laboratory Director
	Day 9 <u>11/09</u>	"	"	"
	Day 10 <u>11/10</u>	"	"	"
	Day 11 <u>11/11</u>	"	"	"
	Day 12 <u>11/12</u>	"	"	"
	Day 13 <u>11/13</u>	"	"	"
	Day 14 <u>11/14</u>	"	"	"
	Day 15 <u>11/15</u>	"	"	"
	Day 16 <u>11/16</u>	"	"	"
	Day 17 <u>11/17</u>	"	"	"
	Day 18 <u>11/18</u>	"	"	"
	Day 19 <u>11/19</u>	"	"	"
	Day 20 <u>11/20</u>	"	"	"
	<u>11/20</u>	"	"	"

37. At end of 20-day testing period, sieve sediment in each aquarium. Count live animals.

Note sublethal responses. Depurate surviving clams in ASW for 48 hr and preserve for bioaccumulation study.

Table B1. Results of solid phase bioassay with the amphipod Rhepoxynius abronius^a

Treatment	Repli- cate	Number of amphipods emerged from sediment (day of exposure)												Number of survivors after 10-day exposure	Number of survivors unable to rebury
		0	1	2	3	4	5	6	7	8	9	10			
Control sediment	1	0	0	0	0	0	0	0	3	2	1	1	19	0	
	2	1	0	0	0	0	0	0	0	1	1	0	18	0	
	3	0	0	0	1	0	0	0	0	0	1	0	18	0	
	4	0	0	0	0	1	0	0	1	0	0	0	19	0	
	5	0	1	0	0	0	0	0	0	0	0	0	19	0	
Reference sediment	1	1	1	1	0	0	0	0	1	1	1	2	20	0	
	2	1	0	0	0	0	0	0	0	0	0	0	17	0	
	3	0	0	0	0	0	0	0	0	0	0	0	20	0	
	4	0	2	1	3	1	0	0	0	0	0	0	17	0	
	5	0	0	1	0	0	0	0	0	0	0	0	18	0	
Alcatraz Disposal Site sediment	1	2	0	0	0	0	0	0	0	1	0	0	14	1	
	2	0	1	0	1	0	0	0	0	0	1	0	18	1	
	3	2	0	0	0	0	0	0	0	0	0	0	19	0	
	4	2	0	0	0	0	0	0	0	0	1	0	15	2	
	5	1	0	0	0	0	0	0	0	0	0	0	14	1	

^aBioassay (10-day test) was conducted at $17.5 \pm 2^\circ\text{C}$ in 1-liter glass beakers under static conditions. Constant illumination was provided throughout the exposure period. Salinity was maintained at 30.5 ppt.

Table B2. Temperature, pH and dissolved oxygen concentration measured during solid phase bioassay with the amphipod Rhepoxynius abronius^a

Parameter	Treatment	Day of exposure										
		0	1	2	3	4	5	6	7	8	9	10
Temperature (°C)	Control sediment	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0
	Reference sediment	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0
	Alcatraz Disposal	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0	17.0
	Site sediment											
pH	Control sediment	7.9	8.1	8.1	8.1	8.0	8.0	8.0	8.1	8.1	8.0	8.0
	Reference sediment	8.0	8.1	8.1	8.1	8.1	8.1	7.9	8.0	8.0	8.1	8.0
	Alcatraz Disposal	7.9	8.1	8.1	8.0	8.0	8.0	8.0	8.0	8.1	8.1	7.8
	Site sediment											
Dissolved oxygen (mg/l)	Control sediment	6.4	8.2	8.1	7.5	7.6	7.7	7.8	7.4	7.7	7.9	8.0
	Reference sediment	6.4	8.3	8.0	7.7	7.6	7.8	7.8	7.5	7.9	7.8	8.0
	Alcatraz Disposal	6.2	8.2	8.2	7.7	7.5	7.8	7.8	7.5	7.9	7.8	8.1
	Site sediment											

^aTemperature, pH, and dissolved oxygen concentrations were measured in beakers established specifically for water-quality determination (conditions were identical to test exposures, but amphipods were absent from the beakers).

Table B3. Results of solid phase bioassays with speckled sanddab (*Citharichthys stigmaeus*) and the clam (*Macoma nasuta*).^a

Treatment	Replicate	Number of survivors ^{b,c}	
		Speckled sanddab	Clam
Control	1	10	10
Sediment	2	10	10
	3	9	10
	4	10	10
	5	9	10
	6	10	8
	7	9	10
	8	10	10
	9	8	10
	10	10	10
Reference	1	8	10
Sediment	2	10	10
	3	9	9
	4	9	10
	5	8	10
	6	9	10
	7	10	10
	8	9	10
	9	9	10
	10	9	10
Alcaltraz	1	9	10
Disposal Site	2	10	10
Sediment	3	9	10
	4	9	9
	5	10	10
	6	10	10
	7	9	10
	8	10	10
	9	10	10
	10	7	10

^aBioassays (20-day tests) were conducted at $17.5 \pm 2^\circ\text{C}$ in 38-liter aquaria. Organisms were exposed to each replicate of a treatment in a single aquarium. Water in aquaria was exchanged by the flow-through method and was aerated. A 14-hr light and 10-hr dark photoperiod was maintained with cool-white fluorescent bulbs. Salinity was maintained at 30.5 ppt.

^bTen (10) individuals of each species were initially exposed to each replicate of a treatment. Thus, a total of 20 animals was employed in each aquarium.

^cIn addition to monitoring survival of both species, burrowing behavior of clams and general behavior of fish was noted each day. No differences were observed among aquaria.

Table B4. Dissolved oxygen concentration measured during solid phase bioassays with speckled sanddab (*Citharusichthys stigmmaeus*) and the clam (*Macoma nasuta*)

		Dissolved oxygen (mg/l)																					
Treatment	Repli- cate	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10	Day 11	Day 12	Day 13	Day 14	Day 15	Day 16	Day 17	Day 18	Day 19	Day 20	
Control sediment	1	7.2	7.8	7.8	7.8	7.8	7.1	6.7	6.9	7.1	7.8	7.6	7.4	7.7	7.6	7.4	7.9	7.8	7.2	7.4	7.5	7.6	
	2	7.2	8.0	7.8	7.6	7.7	6.6	6.4	7.1	7.0	7.7	7.6	7.6	7.6	7.5	7.6	7.8	7.7	7.5	7.4	7.7	7.6	
	3	7.4	8.0	7.9	7.7	7.6	7.3	6.3	6.9	7.3	7.6	7.8	7.6	7.6	7.6	7.6	7.7	7.7	7.4	7.3	7.7	7.5	
	4	7.2	7.8	7.8	7.6	7.7	6.5	6.5	7.0	7.4	7.5	7.6	7.7	7.5	7.6	7.0	7.8	7.8	7.4	7.5	7.5	7.2	
	5	7.4	7.9	8.0	7.6	7.7	6.8	6.7	6.9	7.5	7.5	7.4	7.4	7.7	7.7	7.3	7.8	7.9	7.7	7.5	7.4	7.4	
	6	7.2	7.8	8.0	7.8	7.7	6.9	6.7	6.9	7.5	7.2	7.4	7.7	7.5	7.6	7.0	7.7	7.8	7.9	7.7	7.7	7.4	
	7	7.3	7.8	8.0	7.8	7.8	6.1	6.4	6.9	7.4	7.4	7.5	7.5	7.4	7.5	7.1	7.6	7.9	7.4	7.7	7.4	7.5	
	8	7.2	7.3	8.1	7.8	7.8	6.9	6.6	6.8	7.2	7.5	7.4	7.6	7.5	7.5	7.2	7.6	7.8	7.3	7.5	7.6	7.5	
	9	7.1	7.7	7.9	7.6	7.6	6.4	6.8	6.8	7.5	7.5	7.4	7.6	7.5	7.4	7.2	7.5	7.7	7.5	7.5	7.6	7.5	
	10	7.3	8.0	7.9	7.7	7.6	6.5	6.4	7.0	7.5	7.6	7.6	7.5	7.4	7.4	7.4	7.0	7.8	7.8	7.6	7.6	7.4	
Reference sediment	1	7.1	7.8	8.1	7.8	7.8	7.0	6.9	7.1	7.1	7.5	7.7	7.6	7.4	7.5	7.5	7.8	7.9	7.6	7.7	7.5	7.5	
	2	7.1	8.0	8.1	7.9	7.7	6.8	6.8	7.1	7.2	7.4	7.6	7.7	7.7	7.6	6.9	8.0	7.9	7.5	7.7	7.7	7.4	
	3	7.0	8.0	8.0	7.9	7.7	7.0	6.7	6.9	7.0	7.4	7.4	7.4	7.4	7.6	7.2	7.8	7.9	7.8	7.7	7.7	7.7	
	4	7.3	8.0	8.2	8.0	7.8	6.9	6.7	6.9	7.1	7.2	7.4	7.5	7.4	7.4	7.2	7.7	8.0	7.4	7.5	7.5	7.6	
	5	7.4	7.8	7.9	7.7	7.8	6.7	6.8	7.0	7.3	7.3	7.3	7.4	7.4	7.5	7.0	7.6	7.9	7.7	7.7	7.6	7.5	
	6	7.0	7.7	8.2	7.7	7.7	6.0	6.4	6.9	7.2	7.4	7.4	7.4	7.4	7.6	7.3	7.7	7.9	7.7	7.8	7.6	7.5	
	7	7.2	8.0	8.0	8.0	7.8	6.4	6.4	7.1	7.1	7.4	7.5	7.6	7.6	7.6	7.2	7.8	7.9	7.5	7.6	7.6	7.5	
	8	7.4	7.9	7.9	8.0	7.8	6.8	6.3	6.9	7.4	7.2	7.5	7.7	7.4	7.3	7.2	7.8	7.8	7.7	7.7	7.5	7.7	
	9	7.0	7.9	7.9	7.9	7.8	7.0	6.8	6.8	7.1	7.7	7.5	7.6	7.6	7.4	6.9	7.8	7.9	7.6	7.7	7.7	7.6	
	10	7.1	7.1	7.8	8.0	7.8	7.1	6.8	7.0	7.0	7.5	7.6	7.5	7.7	7.6	7.1	7.8	7.9	7.6	7.7	7.5	7.7	
Alcatraz Disposal Site sediment	1	7.0	8.0	8.0	7.8	7.7	6.5	6.7	7.0	7.5	7.6	7.6	7.7	7.7	7.6	7.6	7.9	7.8	7.8	7.8	7.6	7.7	
	2	7.2	7.9	7.8	7.8	7.7	7.1	6.7	6.9	7.3	7.6	7.5	7.7	7.7	7.4	7.5	8.0	7.9	7.4	7.4	7.6	7.7	
	3	7.1	8.0	8.0	7.8	7.4	6.2	6.4	7.1	7.3	7.4	7.4	7.6	7.6	7.6	7.1	7.8	7.8	7.4	7.7	7.7	7.8	
	4	7.2	7.7	8.1	7.9	7.8	6.2	6.6	7.0	7.4	7.2	7.5	7.5	7.4	7.6	7.5	7.5	7.8	7.6	7.7	7.7	7.7	
	5	6.9	7.8	7.9	8.0	7.7	6.9	6.3	7.1	7.1	7.3	7.6	7.6	7.6	7.7	7.3	7.8	7.9	7.8	7.8	7.6	7.6	
	6	7.1	7.8	7.9	7.7	7.8	6.5	6.4	7.0	7.2	7.5	7.5	7.6	7.6	7.7	7.4	7.6	7.9	7.7	7.7	7.5	7.8	
	7	7.2	7.7	7.9	7.6	7.7	7.3	6.4	7.0	7.2	7.5	7.5	7.5	7.4	7.6	6.8	7.7	8.0	7.9	7.8	7.6	7.7	
	8	6.9	7.6	7.7	7.7	7.7	6.4	6.5	7.0	7.3	7.4	7.6	7.4	7.5	7.5	7.1	7.7	7.9	7.4	7.8	7.6	7.7	
	9	7.1	7.9	7.8	7.8	7.8	6.6	6.7	6.8	7.2	7.5	7.5	7.5	7.6	7.5	7.4	7.7	7.9	7.4	7.8	7.6	7.6	
	10	7.1	8.0	8.0	7.8	7.7	6.3	6.7	6.8	7.5	7.5	7.5	7.5	7.6	7.6	7.7	6.9	7.6	7.9	7.6	7.7	7.7	

Table B5. pH measured during solid phase bioassays with speckled sanddab (Citharichthys stigmaeus) and the clam (Macoma nasuta)

Treatment	Repli- cate	pH																				
		Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10	Day 11	Day 12	Day 13	Day 14	Day 15	Day 16	Day 17	Day 18	Day 19	Day 20
Control sediment	1	7.9	7.8	7.8	7.9	7.9	7.8	7.4	7.7	7.7	7.8	7.8	7.7	7.7	7.8	7.8	7.7	7.7	7.8	7.8	7.7	7.7
	2	7.9	7.8	7.8	8.0	7.9	7.9	7.4	7.8	7.7	7.7	7.6	7.8	7.8	7.7	7.6	7.7	7.7	7.7	7.8	7.8	7.6
	3	7.9	7.9	7.8	7.9	7.9	7.8	7.7	7.8	7.8	7.7	7.8	7.7	7.8	7.7	7.6	7.6	7.6	7.7	7.8	7.8	7.7
	4	7.8	7.8	7.9	7.9	7.8	7.9	7.6	7.9	7.8	7.7	7.7	7.8	7.7	7.8	7.7	7.7	7.7	7.8	7.7	7.7	7.7
	5	7.8	7.9	7.9	7.9	7.9	7.8	7.5	7.7	7.7	7.7	7.7	7.8	7.6	7.9	7.6	7.7	7.8	7.8	7.7	7.7	7.8
	6	7.8	7.9	7.9	7.8	7.9	7.9	7.6	7.7	7.8	7.7	7.7	7.8	7.7	7.8	7.7	7.7	7.7	7.7	7.6	7.7	7.8
	7	7.8	7.8	7.8	7.8	7.9	7.8	7.4	7.8	7.8	7.7	7.8	7.8	7.8	7.8	7.6	7.7	7.7	7.7	7.8	7.7	7.8
	8	7.8	7.9	7.8	8.0	7.9	7.9	7.4	7.7	7.8	7.7	7.8	7.7	7.8	7.8	7.8	7.7	7.7	7.7	7.7	7.8	7.6
	9	7.9	7.9	7.9	7.8	7.9	7.9	7.7	7.7	7.8	7.8	7.8	7.6	7.7	7.7	7.8	7.7	7.8	7.7	7.8	7.7	7.5
	10	7.9	7.9	7.9	7.9	7.9	7.9	7.5	7.7	7.7	7.8	7.8	7.8	7.7	7.7	7.7	7.7	7.7	7.8	7.8	7.7	7.7
Reference sediment	1	7.9	7.8	7.9	7.9	7.8	7.9	7.8	7.7	7.8	7.7	7.6	7.7	7.7	7.9	7.7	7.7	7.7	7.8	7.8	7.9	7.7
	2	7.8	7.8	7.8	8.0	7.8	7.9	7.6	7.8	7.8	7.7	7.7	7.7	7.7	7.7	7.6	7.7	7.7	7.7	7.8	7.8	7.6
	3	7.8	7.8	7.8	7.9	7.8	7.8	7.5	7.7	7.7	7.8	7.7	7.6	7.7	7.8	7.8	7.8	7.7	7.7	7.7	7.8	7.8
	4	7.8	7.8	7.8	7.8	7.9	7.8	7.5	7.7	7.8	7.8	7.7	7.8	7.7	7.8	7.6	7.6	7.6	7.6	7.8	7.8	7.7
	5	7.9	7.9	7.8	7.9	7.8	7.8	7.5	7.8	7.8	7.7	7.7	7.8	7.8	7.8	7.7	7.6	7.7	7.7	7.8	7.7	7.7
	6	7.9	7.9	7.9	7.8	7.8	7.9	7.4	7.8	7.8	7.7	7.7	7.8	7.8	7.7	7.6	7.8	7.8	7.8	7.7	7.7	7.5
	7	7.8	7.8	7.9	7.8	7.9	7.8	7.6	7.8	7.7	7.8	7.8	7.7	7.8	7.7	7.8	7.8	7.8	7.8	7.8	7.7	7.7
	8	7.8	7.9	7.8	7.8	7.8	7.8	7.4	7.7	7.7	7.8	7.7	7.8	7.6	7.8	7.7	7.7	7.7	7.7	7.7	7.8	7.8
	9	7.9	7.9	7.9	7.9	7.8	7.8	7.5	7.8	7.8	7.7	7.8	7.8	7.8	7.8	7.8	7.8	7.8	7.7	7.9	7.8	7.8
	10	7.9	7.8	7.8	7.9	7.9	7.9	7.3	7.8	7.8	7.7	7.8	7.8	7.7	7.8	7.8	7.7	7.6	7.8	7.8	7.7	7.8
Alcatraz Disposal Site sediment	1	7.9	7.9	7.9	7.9	7.9	7.9	7.4	7.8	7.8	7.7	7.8	7.8	7.7	7.7	7.7	7.6	7.7	7.8	7.8	7.8	7.8
	2	7.8	7.9	7.8	7.9	7.9	7.8	7.4	7.7	7.7	7.8	7.8	7.8	7.7	7.7	7.6	7.6	7.7	7.8	7.7	7.7	7.7
	3	7.9	8.0	7.9	7.8	7.9	7.8	7.4	7.7	7.7	7.8	7.7	7.8	7.9	7.8	7.7	7.6	7.8	7.7	7.8	7.7	7.7
	4	7.8	8.0	7.8	7.9	7.9	7.9	7.5	7.8	7.8	7.7	7.7	7.8	7.7	7.7	7.7	7.7	7.7	7.8	7.9	7.8	7.8
	5	7.8	7.9	7.9	7.9	7.8	7.8	7.6	7.8	7.8	7.7	7.7	7.8	7.8	7.7	7.8	7.8	7.8	7.8	7.9	7.7	7.7
	6	7.8	7.9	7.8	8.0	7.8	7.9	7.5	7.7	7.8	7.6	7.7	7.8	7.7	7.7	7.8	7.7	7.8	7.8	7.8	7.9	7.7
	7	7.9	8.0	7.9	7.9	7.8	7.8	7.7	7.8	7.7	7.8	7.8	7.7	7.9	7.8	7.8	7.7	7.6	7.8	7.8	7.8	7.7
	8	7.8	7.9	7.9	8.0	7.8	7.8	7.7	7.6	7.8	7.7	7.8	7.7	7.8	7.7	7.6	7.6	7.7	7.8	7.8	7.8	7.7
	9	7.9	8.0	7.8	7.9	7.8	7.8	7.6	7.8	7.8	7.7	7.8	7.7	7.8	7.7	7.6	7.7	7.7	7.8	7.7	7.8	7.5
	10	7.8	8.0	7.8	8.0	7.9	7.8	7.4	7.7	7.7	7.7	7.8	7.8	7.8	7.7	7.6	7.7	7.7	7.8	7.7	7.8	7.6

Appendix C1. Quality control information pertaining to bioaccumulation studies

Chemical	Type of quality control information			
	Analysis of control sediment from Manchester, Massachusetts ($\mu\text{g/g}$, dry wt.)	Analysis of duplicate sample of tissue from bioaccumulation test ($\mu\text{g/g}$, wet weight)		Analysis of reagent blank
		Replicate 1	Replicate 2	
Arsenic (As)	<1.1	1.1	1.3	<0.05
Cadmium (Cd)	<0.22	0.019	0.018	<0.010
Chromium (Cr)	<2.2	<0.10	<0.10	<0.10
Copper (Cu)	<1.1	1.6	0.97	<1.0
Lead (Pb)	3.4	0.23	0.20	<0.10
Mercury (Hg)	<0.0044	0.022	0.016	<0.005
Selenium (Se)	<1.1	0.13	<0.10	<0.10
Silver (Ag)	<0.22	0.023	0.029	<0.01
Zinc (Zn)	17	9.9	8.9	<0.20
Aldrin	<0.0001	<0.0001	<0.0001	<0.0001
Dieldrin	<0.0001	<0.0001	<0.0001	<0.0001
Chlordane and related compounds	<0.0001	<0.0001	<0.0001	<0.0001
DDT and derivatives	<0.015	<0.015	<0.0015	<0.015
Endrin	<0.0001	<0.0001	<0.0001	<0.0001
Hexachlorocyclohexane	<0.0001	<0.0001	<0.0001	<0.0001
PCBs	<0.20	<0.20	<0.20	<0.20
Toxaphene	<0.0001	<0.0001	<0.0001	<0.0001
Petroleum hydrocarbons	<0.0001	0.30	0.20	0.10

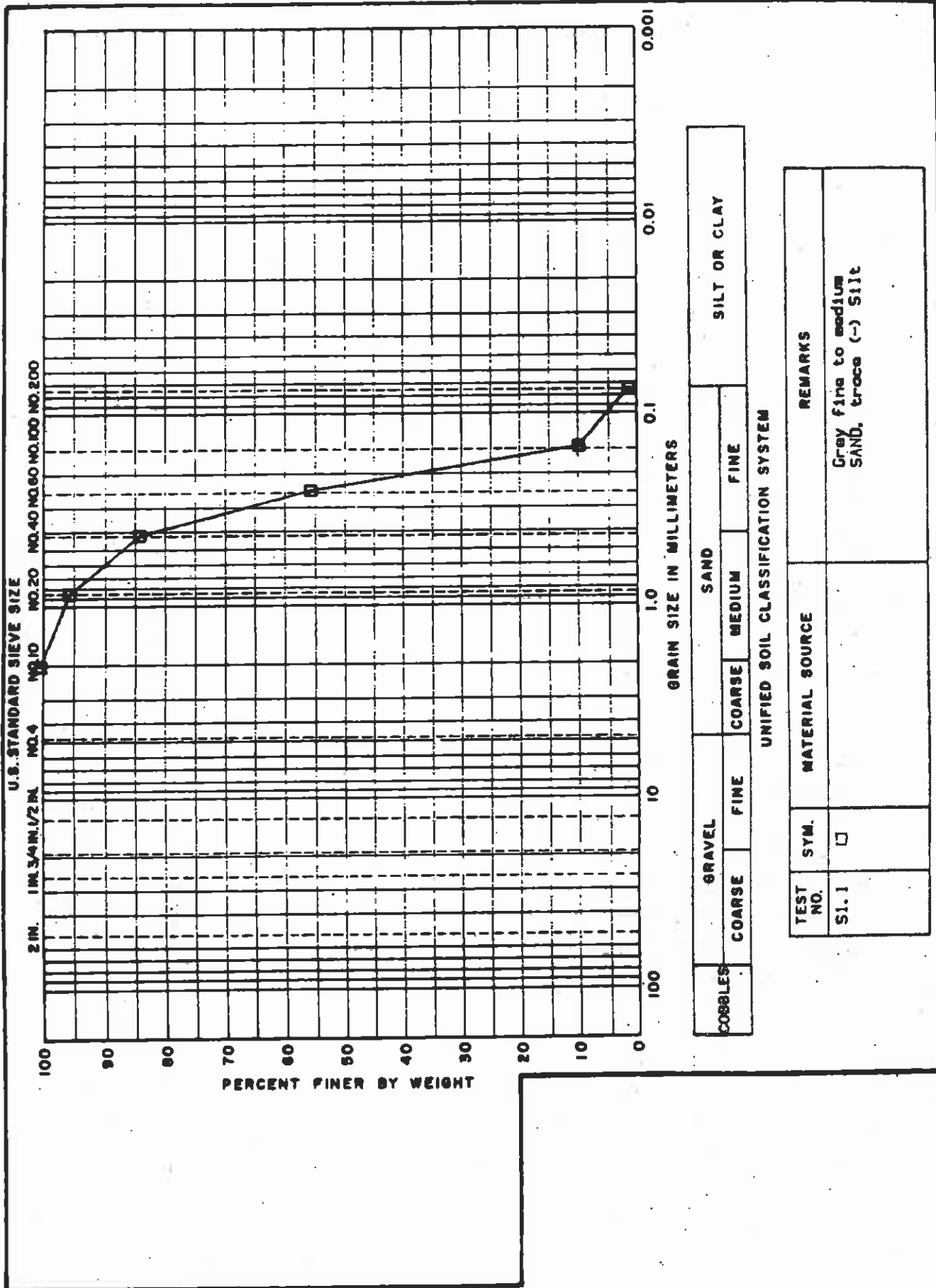


Figure D.1. Grain-size distribution of control sediment collected in Manchester, Massachusetts.

