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Cheol Mo
Virginia Institute of Marine Science

Bruce Neilson
Virginia Institute of Marine Science

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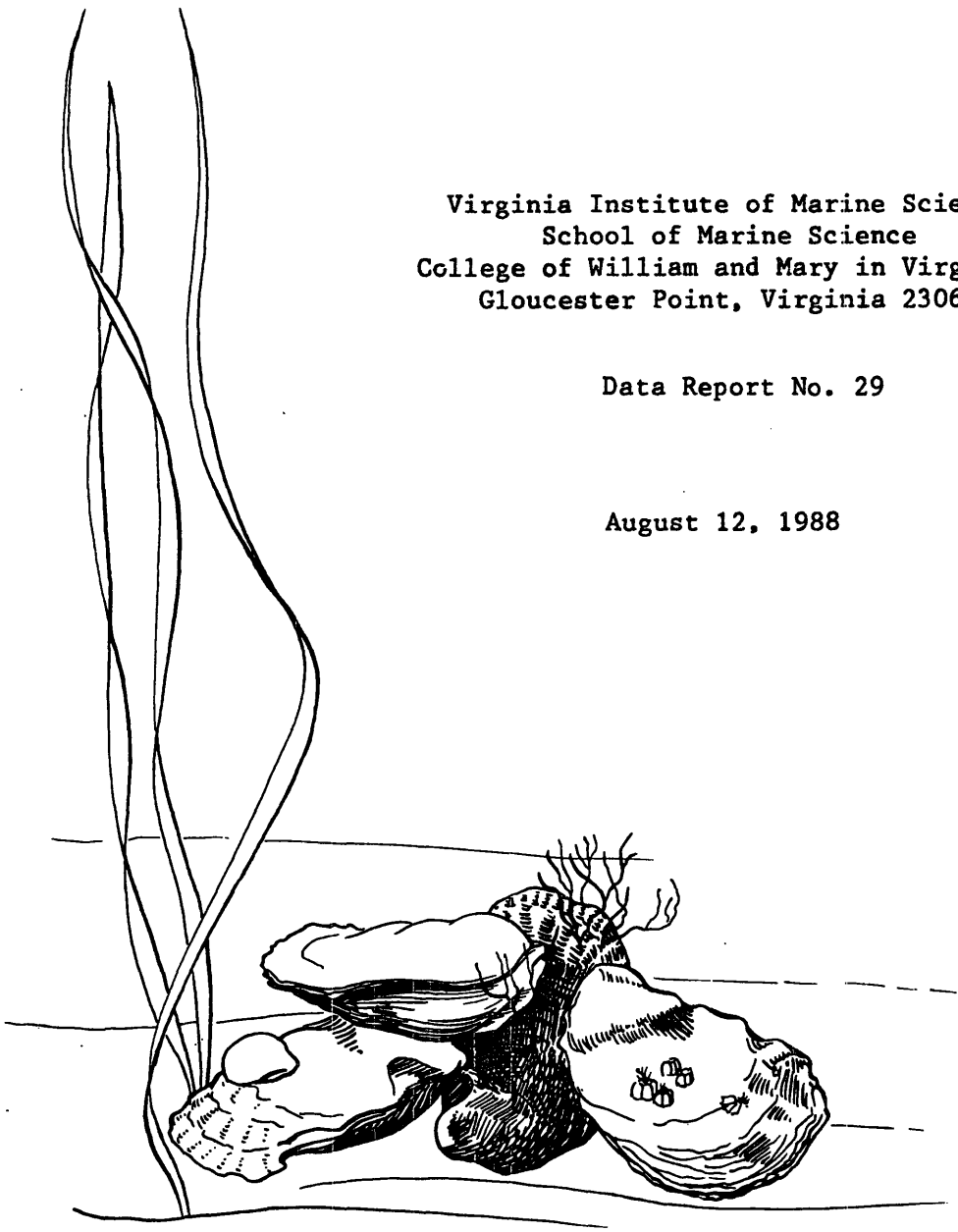

Virginia Institute of Marine Science
School of Marine Science
College of William and Mary in Virginia
Gloucester Point, Virginia 23062

Data Report No. 29

August 12, 1988

Experimental Studies of Zinc-65 Uptake Rates by the American oyster, *Crassostrea virginica* with regard to salinity, sediment concentration, and body size

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PREFACE

These data have been obtained for the calculation of instantaneous uptake rates of zinc by the American oyster, Crassostrea virginica. The uptake rate values are used as a base parameter in a model of heavy metal bio-accumulation in the American oyster. The model will be the main part of the dissertation of Cheol Mo in partial fulfillment for a Ph. D. degree.

Related works which are in preparation are:

(1) "Short term uptake rate of zinc by the American oysters, Crassostrea virginica, - Relationship between body size and metal content."

(2) "Variation of zinc concentrations in oysters related to body size, weight measurement methods, and gut contents."

(3) "Analyses of a model of heavy metal bio-accumulation in the American oyster, Crassostrea virginica - Influence of biological and environmental factors in the bio-accumulation."

The Authors express appreciation for the help of Mr. J. E. Warinner in the measurement of radioactivity.

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ABSTRACT

Three sets of twelve oysters from the James River were placed in three recirculating aquaria dosed with the radioactive tracer zinc-65. All aquaria had the same amount of river bottom sediment which was kept in suspension by the water movement caused by aeration; one aquarium had twice as much tracer as the other two. The salinity of one of the low dose aquaria and the high dose aquarium was maintained at 18 o/oo; the other low dose aquarium was maintained at 12 o/oo. All other factors were kept constant.

Sediment-water-tracer mix was added to the aquaria every 12 hours. Water samples, taken immediately before and after the additions, were filtered with 0.45 micrometer membrane filters. The suspended sediment concentrations and the radioactivities of water and filters were measured.

After 108 hours, the oysters were shucked and the dry weights and the radioactivities measured. Tracer uptake rates were calculated and the relationship between the uptake rate and body size was determined. That relationship was assumed to have the form: uptake equals the product of a constant times weight raised to the power "b" (e.g. a x $\{\text{body size}\}^{b}$). Values for the constants a and b were determined for each aquarium.

INTRODUCTION

Heavy metals in oysters have been studied by many researchers and monitored by regulatory agencies because of the potential hazard to the organism and to human health. Oysters also are often used as a pollution indicator because of their sessile character and because benthic filter feeders tend to accumulate many pollutants, especially heavy metals, to levels many orders higher than in the surrounding waters (Phillips 1977; Warren 1982). However, the relationship between the concentration of metals in the environment and that in oysters has not been clearly defined.

In the natural environment, it can be assumed that the time of exposure is long enough for the organism to be in steady state in terms of uptake and depuration. As with other metal pollution indicator organisms, it is assumed that oysters do not regulate metals to any great extent (Phillips 1977). If the uptake and depuration rates are constant for all sizes of oysters, then a simple linear regression should hold for a given set of physiological and environmental conditions. That is, the concentration in the oyster should be some factor times the ambient concentration.

However, the total concentration of a metal in the environment and that in the organism are not linearly related (Boyden 1974, 1977; Preston 1966) even though some laboratory uptake and depuration studies suggest that the metal bio-concentration of oysters is at equilibrium with the ambient concentration (Romeril 1971). The exponential growth rate of the organism and the dilution effect of tissue mass growth makes

this body size and the body burden per unit mass of tissue relationship complex (Simkiss and Mason 1984; Strong and Luoma 1981; Thomson 1982). Moreover, it has not been understood whether the metal concentration in every cell of the body tissue of oyster changes over the life time or there is a saturation concentration for each cell and the metal concentration of the cell does not increase beyond that concentration (cf. Simkiss and Mason 1984).

Although a better understanding of the metabolic processes of metal uptake and depuration has developed in recent years, many aspects require further study. Information relating body size and metal concentration would be of great help in a variety of applications. For example, differences observed in natural populations could reflect only differences in body size distributions. Management of the resource therefore could be affected by inappropriate interpretation of the data.

The purpose of the present studies is to develop a model of metal uptake and accumulation. The experiments described in this report were intended to determine instantaneous uptake rates for use in the model. These experiments were designed to examine the effect of body size on uptake rate. Surprisingly, there have been few studies of the size-metal burden relationship in American oyster, Crassostrea virginica, other than that of Huggett et al. (1973). In that study, no relationship between body size (wet weight and zinc concentration was found for oysters in the James River. However, samples from different salinity regimes of the river may have been pooled. In a later study (Huggett et al. 1975) it was shown that significant concentration differences were related to salinity. Moreover, the regression of body

weights (which were intentionally selected to be in a narrow range) on metal concentration was determined rather than that of metal concentration on body weights (which should have as wide a range as possible). This could give profoundly different results, especially when it is a Model II regression but one uses the Model I approach. In light of the work by Huggett et al. (1975) and the statistical approach used, the validity of the reported result (i.e. no relationship between body size and zinc concentration) must be questioned.

The problem in studying the metal accumulation in oysters is that the measured metal body burden of oysters from the same site shows a wide variation which makes it difficult, if not impossible, to analyze and interpret the data. Much of the variation is believed due to sampling design, the contribution of the gut content, and the effect of body size. In designing studies, some researchers use wet weight as the measure of body size. The use of wet weight instead of dry weight introduces errors which are relatively large for the smaller organisms. The commonly applied concentrated nitric acid digestion of oyster tissue will release biologically inactive metals which are associated with sediment material in the gut of the organism. Inclusion of metals associated with sediments in the gut can give exaggerated values and/or introduce large variation in results. When there is a relationship between body size and metal concentration, the difference in the distribution of size in a population or among populations will attribute part of the difference in metal concentration.

In most of the previous uptake rate experiments, filtered water was used for the incubation and oysters were intentionally selected to be of

about equal size. Filtered water was used under the assumption that dissolved metal is the major source for metal uptake. Regardless of the validity of that assumption, that method would not give results applicable to oysters in natural environments because oysters are known to detect the absence of particulate material, i.e. food, and to change their behaviour. In particulate-free water, oysters stop pumping water through their gills (Jackim et al. 1977; Jorgensen 1960, 1974, 1975). The results of the earlier uptake studies show a good relationship between environmental concentration and the amount taken up, i.e. a constant uptake rate was observed. However, one must question that relationship given the study designs.

The present study was designed to avoid some of the pitfalls noted above and to provide measurements over a range of body sizes. Suspended river sediments were added to the aquaria, dry weights were used as the measure of body size, and gut contents were removed before the radioactivity and metal content of the oysters was determined. Zinc was chosen because its bio-accumulation by oysters has been extensively studied by many authors, its radioactive isotope (zinc-65) has a relatively long half life (34.4 weeks), it is a physiologically important element, and it has a long biological half life of 300 to 900 days (Seymour and Nelson 1972, 1973; Wolfe 1970).

Oysters of various sizes were held in recirculating tanks and tracer was introduced. Data were collected for individual oysters to obtain a better estimate of kinetic rates. It was assumed that: (1) the newly introduced radioactive tracer was adsorbed to sediment particles and bio-available, (2) depuration was negligible during the

experimental period, (3) the body weight changes of oysters were negligible for the experiment duration, (4) oysters did not discriminate between the radioactive tracer and stable zinc, and (5) there was no adaptive change of uptake rates in the aquaria, at least, for the duration of the experiment.

MATERIALS AND METHODS

Oyster Collection: Three sets of 12 American oysters, Crassostrea virginica, of varying sizes were collected from the mid reaches of the James River near Mulberry Island. Oysters were selected to make the shell length distribution as wide as possible.

Incubation Facility: Three 10-gallon aquaria with racks were filled with sea water and aerated with an air pump.

Sea Water: York River water was filtered through 1 micrometer filters. Distilled-deionized water was added to make 18 and 12 o/oo salinities.

Radioactive Tracer Mix: Bottom sediments from the oyster collection sites in the James River were collected and mixed with filtered York River water. The volume of water was about twice that of the sediments to make sediment particles suspend reasonably freely in the water. The mix was filtered with a 63 micrometer opening sieve. The sediment-water mix was stirred with a glass rod. For ten minutes the relatively fast sinking silt and sand portion of the sediments was allowed to settle to the bottom of the container. The supernate was transferred to another container and the remaining settled sediments were discarded. This procedure was repeated until no discernible amount of sediment particles settled within a ten minute period. Then 5 ml was taken and dried at 105 ^{o}C until there was no significant weight change. This sample was used to determine the nutrient and water contents. The nutrient analyses were made with a Carlo Erba "C.N. Analyzer" (Table 1).

Zinc-65 in the form of chloride ($ZnCl_2$) in hydrochloric acid (HCl) (Table 2) was mixed with the sediment-water mix and left for one day

(cf. Haven et al. 1981; Chleck et al. 1963). Two sets of tracer mix were prepared. One mix had twice as much tracer as the other mix. Every 12 hours, 12.5 ml of tracer mix was added to each aquarium with the goal of maintaining radioactivity concentrations of 1.0 (aquarium no. 1) and 0.5 (aquaria no. 2 and no 3) microcurie per liter with sediment concentrations of approximately 50 mg/L, the nominal value for the near bottom waters in Chesapeake Bay estuaries (Harris et al. 1980; Nichols et al. 1981, 1983).

It was assumed that a chemical equilibrium would be established when the tracer mix was put into sea water, and that the metal was in three forms: free ion, complexed with ligands, and sediment adsorbed. The chemical speciation in the water column, however, was and will be ignored. Only total dissolved zinc will be examined and all of it will be assumed to be biologically available. The effect of the acidity of the mix was determined to be too small to be of concern.

GENERAL LABORATORY EXPERIMENTAL PROCEDURES:

1. Oysters were brushed under running sea water to remove adhering mud and placed in aquaria. Aerators were placed in the aquaria and turned on. Every four hours, the aquaria were drained and refilled with unfiltered York River water. This was done for 7 days. Oysters filtered particulate materials from the water and deposited faecal pellets to the bottom of the aquaria. The faecal pellets were not re-suspended by the aeration. (cf. Haven and Morales-Alamo 1968)

2. Three aquaria were half filled with the prepared York River water (18 o/oo water to aquaria nos. 1 and 2, and 12 o/oo water to

aquarium no. 3). Then 12 ml of the tracer mix were placed in each aquarium (high dose to aquarium no. 1 and low dose to aquaria nos. 2 and 3). Water was added to a final volume of 25 liters. The aeration pump was turned on and the aquaria were left for 24 hours.

3. The initial (t=0) water samples (including suspended particles) were taken from each aquaria and the oysters were placed into the aquaria.

4. Every 12 hours, 12.5 ml of the sediment-tracer mix was added to the aquaria. Before and after the addition, 100 ml water samples were taken. The study continued for four and half a days, a time period which is convenient to compare the results with other researchers' work (cf. Fitzgerald and Skauen 1963; Seymore and Nelson 1973).

5. The aquaria were covered by hard boards. Whenever there was any marked drop of the water level, distilled-deionized water was added to maintain a 25 liter volume and a constant salinity for every aquarium.

6. At the end of the experiment (t=4.5 days), shells of oysters were removed with a stainless steel oyster shucking knife. The tip of a pipette was inserted to the anal opening of each oyster and distilled-deionized water was injected to flush gut contents out the mouth (cf. Galtsoff 1964). The soft body tissues were placed in separate 20 ml polyethylene liquid scintillation counting vials.

7. Above samples were dried at 105 ^{o}C until there was no weight change and weighed again for dry weight measurement.

8. Each dried oyster tissue was crushed with a glass rod and transferred to a vial for the "BioGamma Counter". The liquid

scintillation vial and the glass rod were rinsed with 4 ml of 50% HNO_3 acid and the acid was added to the oyster tissue sample in the counting vial.

Radioactivity Measurement: The "electron capture" accounts for 98% of zinc-65 radioactive decay. Fifty-one percent of the gamma rays produced by its radioactive decay have 1.116 MeV of energy which is not measured efficiently by liquid scintillation spectrometers. A Beckman "BioGamma II", an automated sodium-iodide crystal detector which uses trays of 5 ml counting vials, was used. The following procedures were used:

1. A standard was placed into the counting chamber. Window selector modules were placed into channel 1 slot and channel 2 slot and the minima and maxima were set at 10.0. The standard was counted for 20 seconds. The minima were decreased from 10.0 to 0.0 in increments of 0.5; 20 second counts were made at each step. The counts were graphed (Figure 1) to find the counting peak. As a result, the minimum and maximum for channel 1 were set to 5.0 and 6.5 respectively to obtain counts with minimal noise, and the minimum and maximum for channel 2 were set to 3.0 and 7.0 respectively to count with the most efficiency.

2. 3.5 ml of standard tracer with 0.8364 microcurie/ml radioactivity (total of 2.9274 microcurie) were placed into the counting chamber and counted for 2 minutes. The counts were repeated three times. The results were:

	channel 1	channel 2
counts:	293582, 290186, 296594	531173, 524262, 534374
average:	293154	528269
efficiency	4.5%	8.0%

3. The filters, 4 ml of filtrates, and oyster samples in the vials were placed into the trays and counted for 2 minutes each in both channels.

4. For every 10 samples, a blank sample and a duplicate sample were counted.

5. The calibrated results from channel 1 and channel 2 agree well (see Figure 2). Consequently, the two values were averaged in all subsequent calculations.

Statistical Analysis: The radioactive tracer concentrations of oysters were plotted against the dry weights. Relationships were analysed using "LREG Procedure" of "SAS Statistics" program (SAS Institute Inc. 1985).

RESULTS

The salinities and temperatures of the aquaria were fairly well maintained throughout the experiment period (Table 3). Temperature differences among the aquaria are thought to be the result of drafts.

When the sediment-tracer mix was added to the aquaria, the water became turbid; twelve hours later, the water was almost clear (Figure 3). Little sediment was deposited on the bottom except for faecal pellets. It was easily observed that the oysters were pumping water actively throughout the experiment. Dead oysters were easily recognizable because the shells were open wide but the animal did not pump water. Any dead oysters were removed from the aquaria immediately. At the end of the experiments, 11 oysters were left in each aquarium.

Radioactivity counts were converted to tracer concentrations by multiplying the count by the efficiency calculated from the standard: channel 1 was 2.9274/293154 and channel 2 was 2.9274/528269. Suspended solids concentrations (Table 4) and water concentrations (Table 5) were adjusted to be in a common unit (microcurie per liter).

About one-fourth of the tracer was released into the water phase when the tracer mix was introduced. Although the tracer concentrations in the water and suspended solid phases decreased by about the same amount for each period (Figure 4), the weight specific tracer concentrations of the suspended solids (microcurie per gram dry weight solids) in the aquaria were reduced too (Figure 5). The weight specific radioactivity of the suspended solids in an aquarium without oysters did not change significantly over the course of the experiment. Therefore

it was inferred that an additional portion of the tracer adsorbed to suspended solids was released to the water as the metal concentration (including the tracer) in the water column decreased through uptake by the oysters.

The tracer concentrations for the oyster samples were calculated by multiplying the radioactivity count by the counting efficiency for each counting channel and then dividing the result by the corresponding oyster dry body weight (Table 6). The oyster tracer concentrations, which were the relative amount of uptake for 4.5 days, were transformed. Following Boyden (1974), it has been assumed that total metal concentration per individual (Y) is related to body weight (X) as a power function, or:

$$Y = aX^b$$

$$\text{then } \log Y = \log a + b \log X$$

Accordingly, the tranformation performed in this study was:

$\log_{10}(\text{concentration}) = a + b \{ \log_{10}(\text{body weight}) \}$.

A linear regression line was fitted for each aquarium by the "least square method" {Figures 6(a) to 6(c)} using the log-transformed data set. The results are shown in Figures 6(d) to 6(f) in terms of actual concentration and body weight, along with the transformed linear regression line from the previous figures {Figures 6(a) to 6(c)}.

Table 1. Characteristics of the Sediment Used for the Sediment-water-tracer-mix

Wet weight of 5 ml	:	5.1421 gram
Dry weight of 5 ml	:	0.4814 gram
Solid content	:	0.096 g/ml
Water content	:	90.6%
Carbon content	:	4.353% of dry weight
Nitrogen content	:	0.542% of dry weight

Table 2. Radioactive Tracer Characteristics

Name	:	Zinc-65
Supplier	:	NEV Research Product Division of DuPont
Catalogue No.	:	NEZ-111
Total acitivity of the shipment	:	1.02 mCi
Purity	:	0.99
Specific activity	:	1.97 mCi/mg
Concentration	:	10.0 mCi/ml
Volume	:	0.1 ml
Form	:	$Zn^{*}Cl_2$ in 0.5 M HCl

Table 3(a). Water and Suspended Solids in Aquarium No. 01

	TIME (day)	SALINITY (o/oo)	TEMP. (°C)	pH	eH	S.S. (dry-g/L)
a	0.0	17.30	23.4	7.12	504.8	0.058
b	0.5	17.63	19.5	7.37	487.5	0.031
a	0.5		21.4	7.42	474.6	0.048
b	1.0	17.68	19.7	7.46		0.029
a	1.0	17.53	19.8	7.45		0.072
b	1.5		19.4	7.59		0.032
a	1.5	17.43	22.8	7.45		0.082
b	2.0	17.46	22.4	7.45		0.040
a	2.0		22.3	7.50		0.078
b	2.5	17.32	22.2	7.52		0.042
a	2.5	17.27	19.6	7.47		0.085
b	3.0		20.2	7.38		0.037
a	3.0	17.57	23.5	7.50		0.094
b	3.5	17.10	22.5	7.46		0.041
a	3.5		23.0	7.47	489.5	0.077
b	4.5		25.1	7.59	480.0	0.035

a (b) = after (before) tracer mix added to aquarium.

Table 3(b). Water and Suspended Solids in Aquarium No. 02

	DAY (day)	SALINITY (o/oo)	TEMP. (°C)	pH	eH	S.S. (dry-g/L)
a	0.0	17.20	24.4	7.40	468.7	0.063
b	0.5		19.9	7.34	463.0	0.036
a	0.5	17.73	21.4	7.50	461.0	0.063
b	1.0		21.2	7.38		0.038
a	1.0	17.86	20.0	7.43		0.080
b	1.5	17.72	21.6	7.40		0.037
a	1.5	17.69	22.3	7.47	480.0	0.089
b	2.0	17.74	21.3	7.55		0.045
a	2.0		21.4	7.45		0.087
b	2.5	17.32	20.9	7.51		0.036
a	2.5		20.8	7.62		0.091
b	3.0	17.34				0.038
a	3.0	17.20	21.4	7.51		0.078
b	3.5		22.5	7.46	489.5	0.041
a	3.5		21.0	7.49	482.7	0.075
b	4.5		23.1	7.54	488.0	0.039

a (b) = after (before) tracer mix added to aquarium.

Table 3(c). Water and Suspended Solids in Aquarium No. 03

	DAY (day)	SALINITY (o/oo)	TEMP. (^{o}C)	pH	eH	S.S. (dry-g/L)
a	0.0	12.73	22.7	7.47	480.0	0.056
b	0.5	13.07	20.2	7.16	471.0	0.024
a	0.5	13.02	23.7	7.25	488.0	0.044
b	1.0	13.01	20.4	7.35		0.022
a	1.0	12.01	20.3	7.38		0.068
b	1.5	12.93	20.9	7.55	480.0	0.026
a	1.5	12.81	23.2	7.49		0.079
b	2.0	12.89	22.0	7.49		0.026
a	2.0	12.65	21.6	7.45		0.071
b	2.5	12.76	20.9	7.49		0.030
a	2.5	12.72	21.4	7.47		0.085
b	3.0	12.69	21.3	7.42		0.037
a	3.0	12.48	22.0	7.51		0.077
b	3.5	12.51	23.3	7.45	502.2	0.019
a	3.5	12.10	23.2	7.43	483.0	0.058
b	4.5		23.3	7.54	488.0	0.026

a (b) = after (before) tracer mix added to aquarium.

Table 4(a). Zinc-65 Radioactivity of Suspended Solids in Aquarium No. 01

	TIME (day)	S.S. CONC. (gram/liter) (dry weight)	COUNT* Channel 1	COUNT* Channel 2	RADIOACTIVITY (microcuri per) (liter	RADIOACTIVITY (microcuri per) gram-s.s.)
a	0.0	0.058	968	1832	0.0991	1.7085
b	0.5	0.031	130	267	0.0139	0.4480
a	0.5	0.048	4015	7037	0.3954	8.2384
b	1.0	0.029	351	712	0.0373	1.2846
a	1.0	0.072	4431	8307	0.4514	6.2695
b	1.5	0.032	348	699	0.0367	1.1482
a	1.5	0.082	5669	10578	0.5761	7.0261
b	2.0	0.040	727	1340	0.0734	1.8357
a	2.0	0.078	6065	11155	0.6119	7.8449
b	2.5	0.042	802	1550	0.0830	1.9760
a	2.5	0.085	7634	13838	0.7646	8.9950
b	3.0	0.037	560	1075	0.0577	1.5607
a	3.0	0.094	4563	15065	0.6452	6.8643
b	3.5	0.041	279	1075	0.0437	1.0662
a	3.5	0.077	7737	14098	0.7769	10.0899
b	4.5	0.035	240	561	0.0275	0.7865

* 100 ml of water filtered through 0.45 um filter.

Table 4(b). Zinc-65 Radioactivity of Suspended Solids in Aquarium No. 02

	TIME (day)	S.S. CONC. (gram/liter) (dry weight)	COUNT* Channel 1	COUNT* Channel 2	RADIOACTIVITY (microcurie per) (liter	RADIOACTIVITY (microcurie per) gram-s.s.)
a	0.0	0.063	968	1320	0.0849	1.3477
b	0.5	0.036	111	269	0.0130	0.3610
a	0.5	0.063	1752	3235	0.1771	2.8113
b	1.0	0.038	247	503	0.0263	0.6913
a	1.0	0.080	3549	6508	0.3575	4.4690
b	1.5	0.037	321	663	0.0344	0.9297
a	1.5	0.089	3762	7063	0.3835	4.3094
b	2.0	0.045	598	1100	0.0603	1.3408
a	2.0	0.087	6167	11155	0.6170	7.0919
b	2.5	0.036	441	842	0.0453	1.2597
a	2.5	0.091	4881	9100	0.4958	5.4488
b	3.0	0.038	516	1054	0.0550	1.4465
a	3.0	0.078	5631	10622	0.5755	7.3777
b	3.5	0.041	320	664	0.0344	0.8384
a	3.5	0.075	2986	5445	0.3000	3.9994
b	4.5	0.039	343	695	0.0364	0.9329

* 100 ml sample filtered through 0.45 um filter.

Table 4(c). Zinc-65 Radioactivity of Suspended Solids in Aquarium No. 03

	TIME (day)	S.S. CONC. (gram/liter) (dry weight)	COUNT* Channel 1	COUNT* Channel 2	RADIOACTIVITY (microcurie per) (liter	RADIOACTIVITY (microcurie per) gram-s.s.)
a	0.0	0.056	950	1773	0.0966	1.7243
b	0.5	0.024	107	245	0.0121	0.5054
a	0.5	0.044	1012	1907	0.1034	2.3492
b	1.0	0.022	292	545	0.0297	1.3491
a	1.0	0.068	2531	4598	0.2538	3.7319
b	1.5	0.026	301	637	0.0327	1.2569
a	1.5	0.079	3498	6464	0.3538	4.4779
b	2.0	0.026	447	810	0.0448	1.7216
a	2.0	0.071	5602	9821	0.5518	7.7721
b	2.5	0.030	322	651	0.0341	1.1372
a	2.5	0.085	4329	8052	0.4392	5.1676
b	3.0	0.037	560	1075	0.0577	1.5607
a	3.0	0.077	4744	8872	0.4827	6.2686
b	3.5	0.019	205	459	0.0230	1.2081
a	3.5	0.058	2782	5283	0.2853	4.9187
b	4.5	0.026	220	476	0.0242	0.9297

* 100 ml of water filtered through 0.45 um filter.

Table 5(a). Zinc-65 Radioactivitiy of Water in Aquarium No. 01

	TIME (day)	COUNT* Channel 1	COUNT* Channel 2	RADIOACTIVITY (microcurie/liter)
a	0.0	629	1201	1.6171
b	0.5	516	1007	1.3416
a	0.5	786	1523	2.0361
b	1.0	589	1072	1.4778
a	1.0	760	1426	1.9364
b	1.5	536	1027	1.3804
a	1.5	852	1638	2.1981
b	2.0	554	1073	1.4348
a	2.0	793	1463	2.0033
b	2.5	592	1106	1.5051
a	2.5	845	1584	2.1520
b	3.0	633	1191	1.6151
a	3.0	825	1576	2.1215
b	3.5	547	1069	1.4233
a	3.5	784	1464	1.9927
b	4.5	387	956	1.1453

* Count made using 4 ml of filtered water.

Table 5(b). Zinc-65 Radioactivity of Water in Aquarium No. 02

	TIME (day)	COUNT* Channel 1	COUNT* Channel 2	RADIOACTIVITY (microcurie/liter)
a	0.0	438	869	1.1487
b	0.5	406	788	1.0526
a	0.5	530	996	1.3515
b	1.0	465	938	1.2302
a	1.0	549	1070	1.4265
b	1.5	490	934	1.2586
a	1.5	614	1168	1.5755
b	2.0	484	959	1.2684
a	2.0	793	1463	2.0033
b	2.5	451	871	1.1663
a	2.5	602	1141	1.5418
b	3.0	500	923	1.2635
a	3.0	564	1071	1.4459
b	3.5	485	931	1.2503
a	3.5	539	1087	1.4257
b	4.5	427	845	1.1183

* Count made on 4 ml of filtered water.

Table 5(c). Zinc-65 Radioactivity of Water in Aquarium No. 03

	TIME (day)	COUNT* Channel 1	COUNT* Channel 2	RADIOACTIVITY (microcurie/liter)
a	0.0	415	822	1.0874
b	0.5	393	745	1.0066
a	0.5	476	941	1.2460
b	1.0	421	803	1.0817
a	1.0	552	1018	1.3942
b	1.5	442	829	1.1260
a	1.5	507	1014	1.3352
b	2.0	398	719	0.9948
a	2.0	565	1062	1.4409
b	2.5	402	834	1.0795
a	2.5	537	992	1.3574
b	3.0	389	756	1.0092
a	3.0	500	973	1.2981
b	3.5	378	730	0.9775
a	3.5	465	923	1.2198
b	4.5	349	704	0.9233

* Count made with 4 ml of filtered water.

Table 6(a). Zinc-65 Radioactivity of the Oysters in Aquarium No. 1

ID	WEIGHT (dry-gram)	COUNT* (numbers/2 min.) ch. 1	ch. 2	RADIOACTIVITY (microcurie microcurie/g-wt.) total	specific
A12	0.1174	39420	71839	0.4161	3.5443
A15	0.1389	200760	361001	2.1050	15.1545
A24	0.1521	168869	305016	1.7746	11.6670
A16	0.3498	117310	213749	1.2382	3.5397
A04	0.3642	150874	273596	1.5886	4.3619
A11	0.6714	98037	176164	1.0276	1.5305
A02	0.7197	464836	812705	4.8062	6.6781
A23	0.7828	266597	476936	2.7881	3.5617
A21	0.9936	267726	481916	2.8086	2.8266
A06	1.0092	132847	237701	1.3894	1.3768
A14	1.3452	312904	566459	3.2919	2.4471

Number of oysters: 11

Uptake equation: $\log Y = 0.393150 + (-0.580395) * \log X$

* Count made using whole soft body tissue.

Table 6(b). Zinc-65 Radioactivity of the Oysters in Aquarium No. 2

ID	WEIGHT (dry-gram)	COUNT* (numbers/2 min.) ch. 1	ch. 2	RADIOACTIVITY (microcurie microcurie/g-wt.) total	specific
A09	0.1691	93801	174561	1.0007	5.9178
A01	0.2801	56791	103191	0.5986	2.1370
A19	0.2929	111142	199629	1.1647	3.9763
A05	0.2994	97743	155566	0.9659	3.2261
A10	0.4706	65581	118952	0.6906	1.4675
A13	0.4867	118184	215089	1.2467	2.5615
A07	0.6656	117147	211865	1.2318	1.8507
A27	0.7659	80662	145689	0.8476	1.1067
A17	0.7807	80818	146200	0.8499	1.0887
A28	0.8338	143394	257775	1.5033	1.8029
A08	1.0897	122377	218718	1.2792	1.1739

Number of oysters: 11

Uptake equation: log Y = 0.052273 + (-0.832761) * log X

* Count made using whole soft body tissue.

Table 6(c). Zinc-65 Radioactivity of the Oysters in Aquarium No. 3

ID	WEIGHT (dry-gram)	COUNT* (numbers/2 min.)		RADIOACTIVITY (microcurie microcurie/g-wt.)	
		ch. 1	ch. 2	total	specific
A22	0.1968	59787	107886	0.6243	3.1723
A03	0.2711	40895	74175	0.4306	1.5885
A40	0.2809	125062	224207	1.3093	4.6611
A35	0.3219	18484	104868	0.6123	1.9023
A29	0.3535	74380	134442	0.7819	2.2119
A30	0.6077	1141703	254340	1.4844	2.4426
A38	0.6289	187995	343709	1.9876	3.1605
A31	0.8028	164412	281921	1.6838	2.0974
A36	0.8227	126222	227207	1.3241	1.6095
A32	0.9309	61809	112084	0.6508	0.6991
A33	1.1285	86428	156058	0.9081	0.8047

Number of oyster: 11

Uptake equation: $\log Y = 0.102598 + (-0.607743) * \log X$

* Count made using whole soft body tissue.

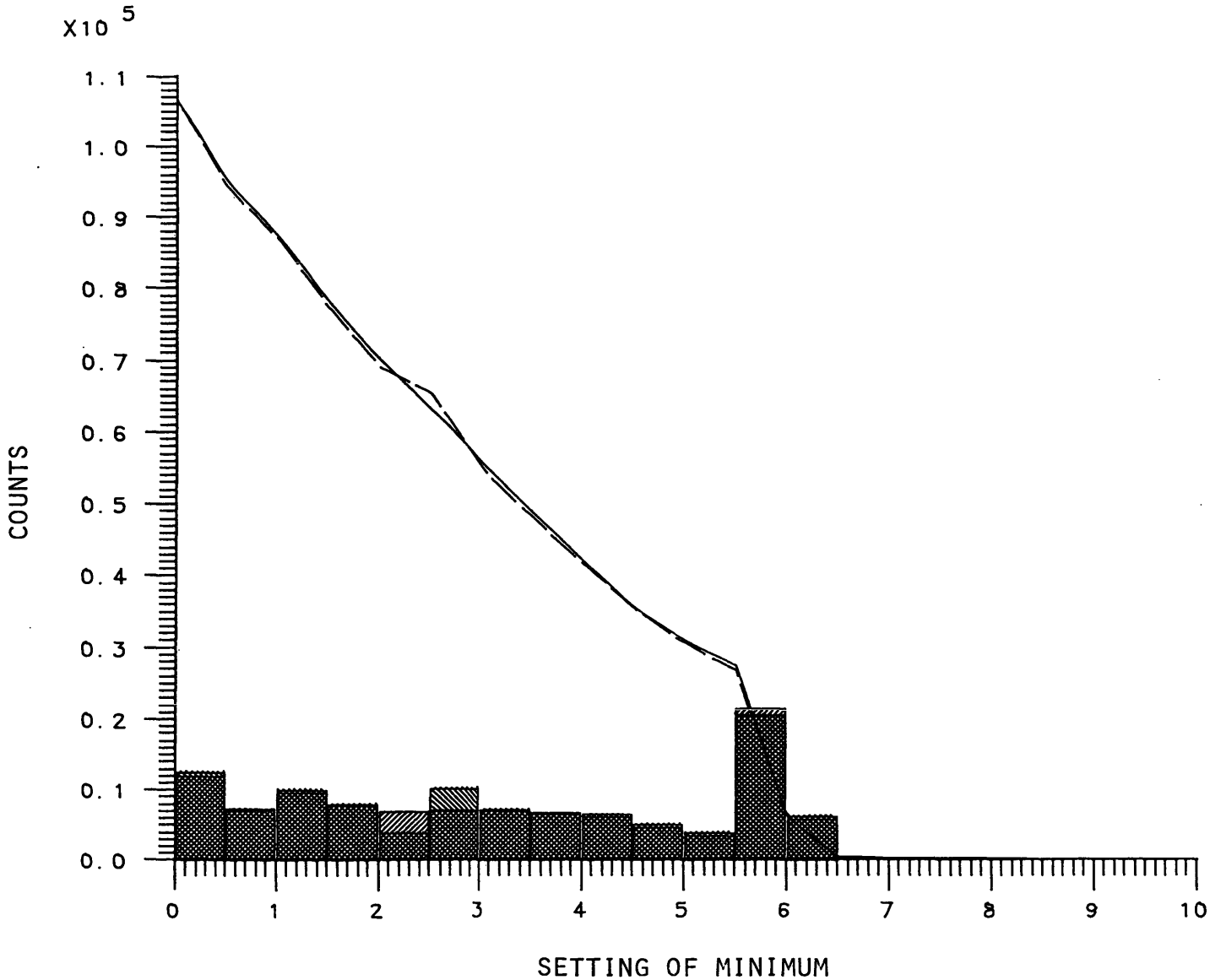

Figure 1. Counting efficiency for the gamma ray detector. Counts resulting from radioactive decay of a standard were measured for 20 seconds on two channels of a gamma ray detector. The maximum setting was held constant at 10.0 and the minimum setting reduced from 10.0 to 0.0 in 0.5 unit decrements. Both counts and cumulative counts are shown.

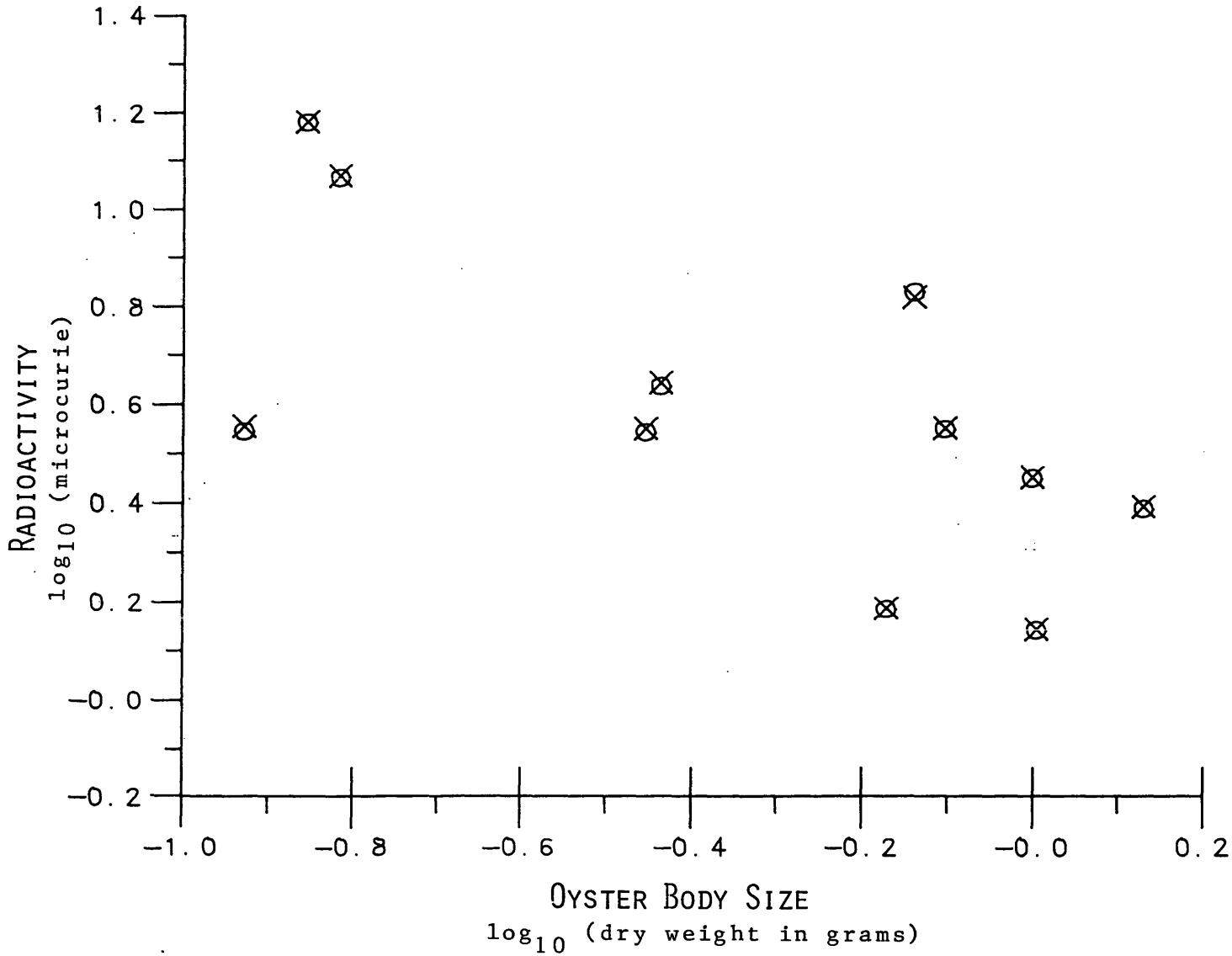

Figure 2(a). Comparison of radioactivity measurements from 2 channels for oysters in aquarium No. 1. Counts were normalized with readings from a standard.

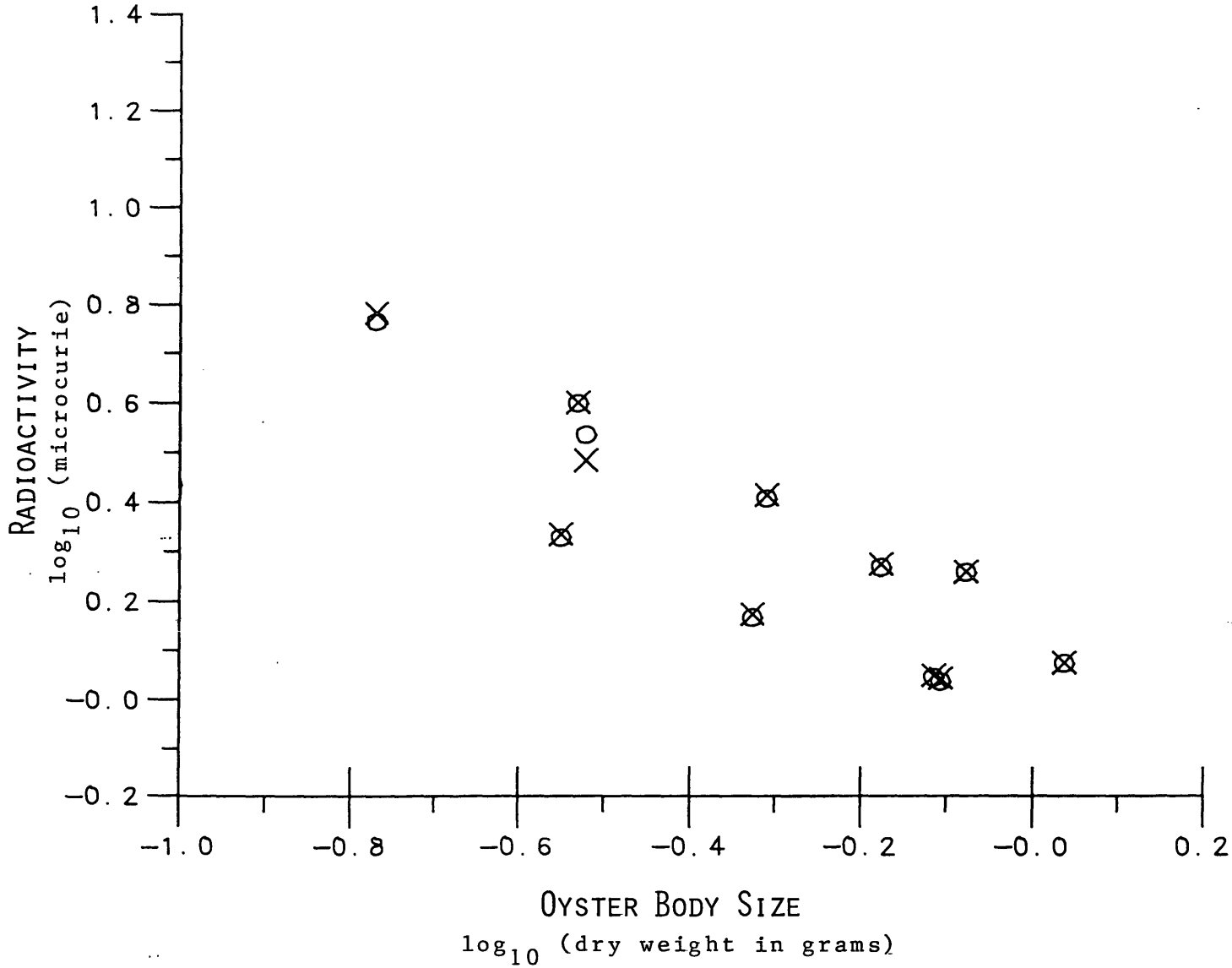

Figure 2(b). Comparison of radioactivity measurements from 2 channels for oysters in aquarium No. 2. Counts were normalized with readings from a standard.

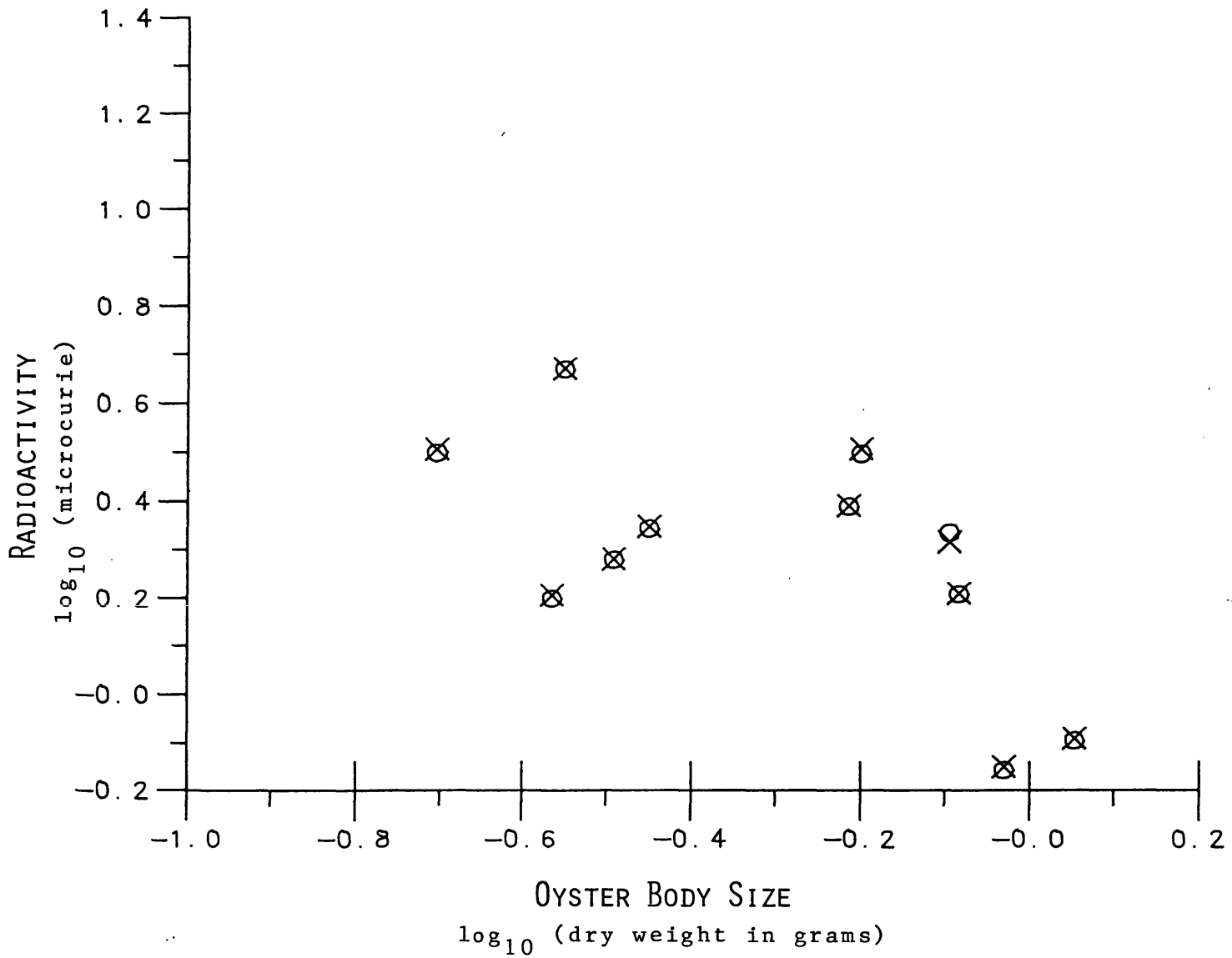

Figure 2(c). Comparison of radioactivity measurements from 2 channels for oysters in aquarium No. 3. Counts were normalized with readings from a standard.

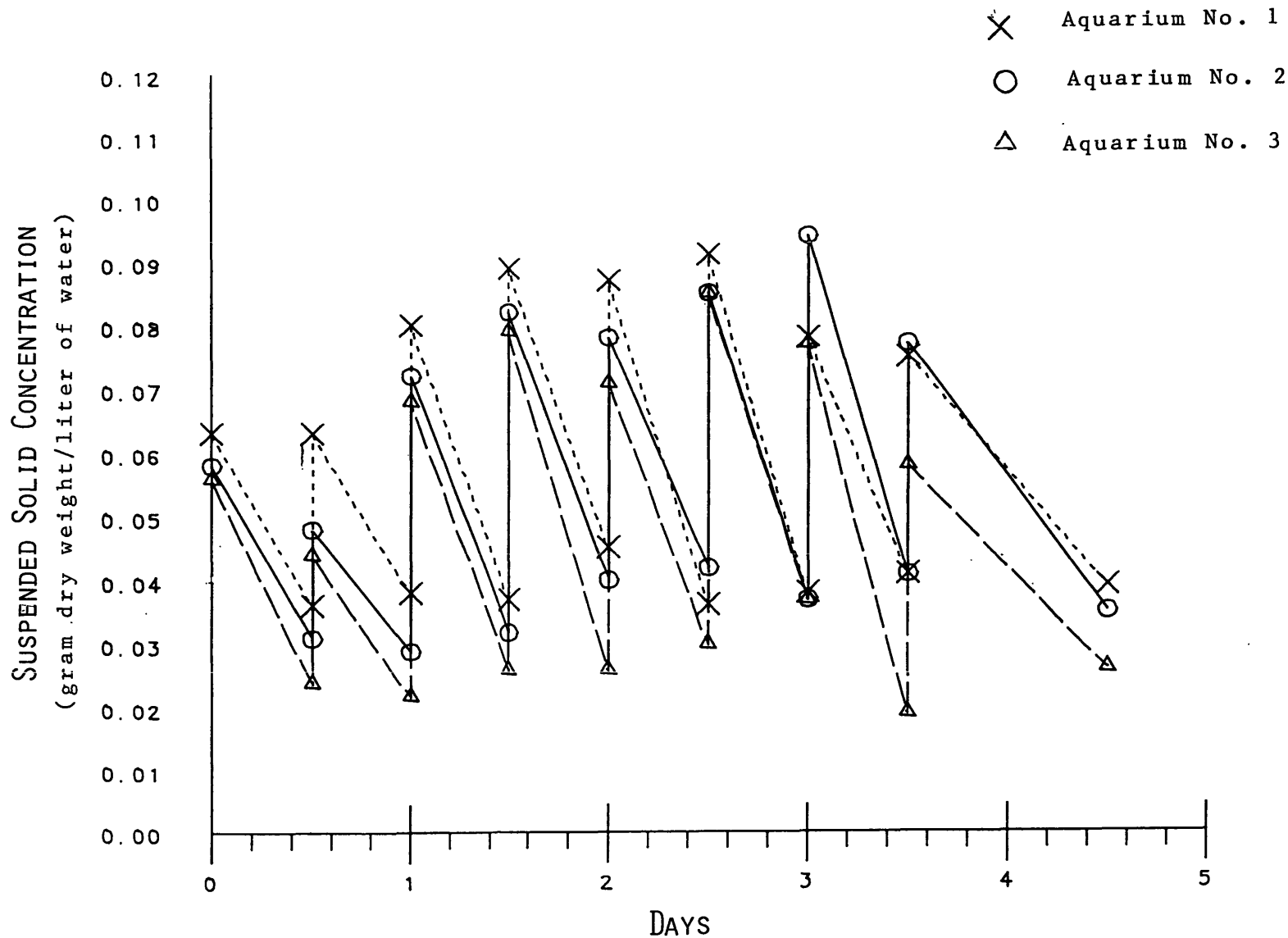

Figure 3. Time variation of suspended solids concentrations in the three aquaria.

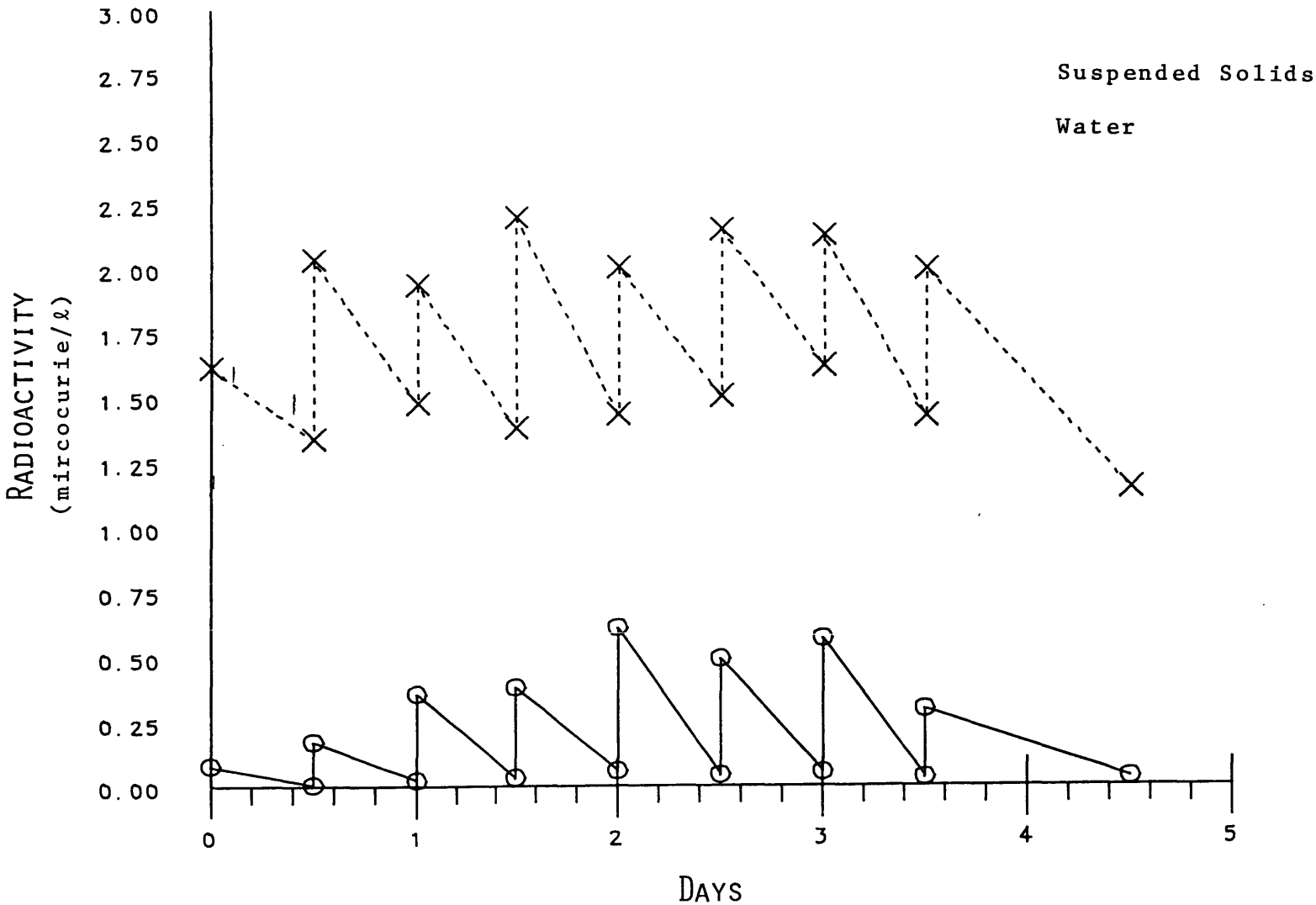

Figure 4(a). Time variation of radioactivity of suspended solids and water in aquarium No. 1.

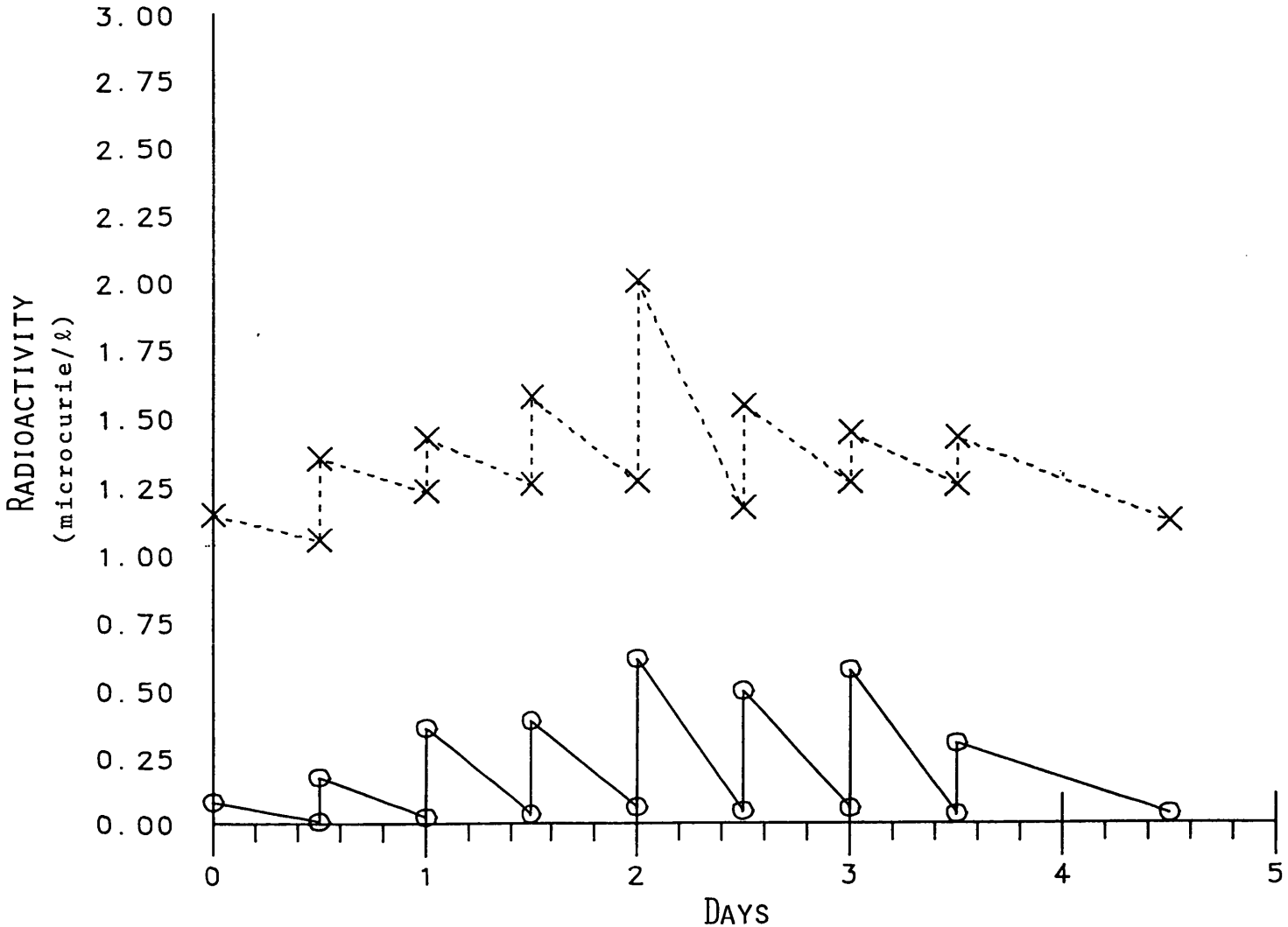

Figure 4(b). Time variation of radioactivity of suspended solids and water in aquarium No. 2.

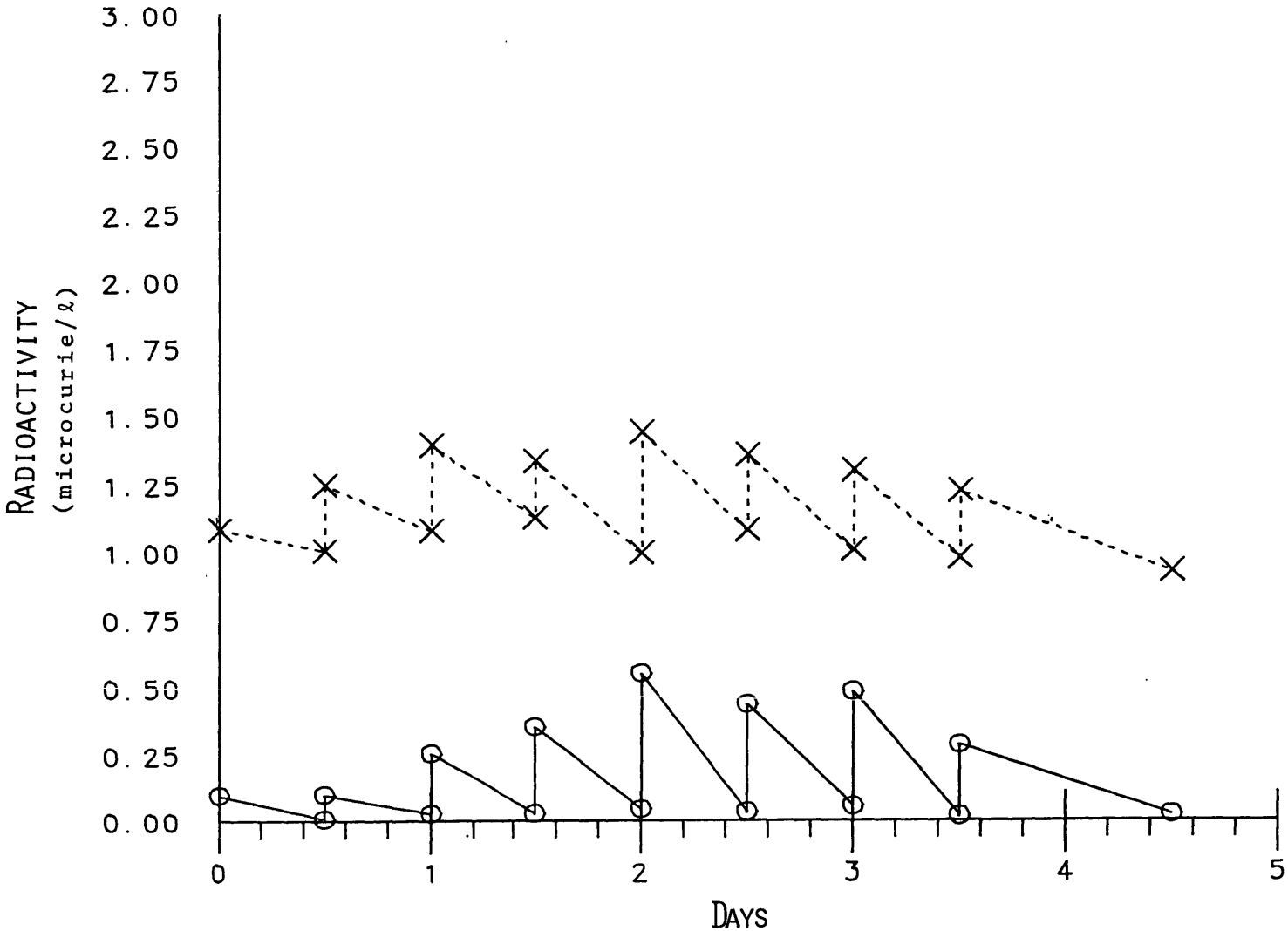

Figure 4(c). Time variation of radioactivity of suspended solids and water in aquarium No. 3.

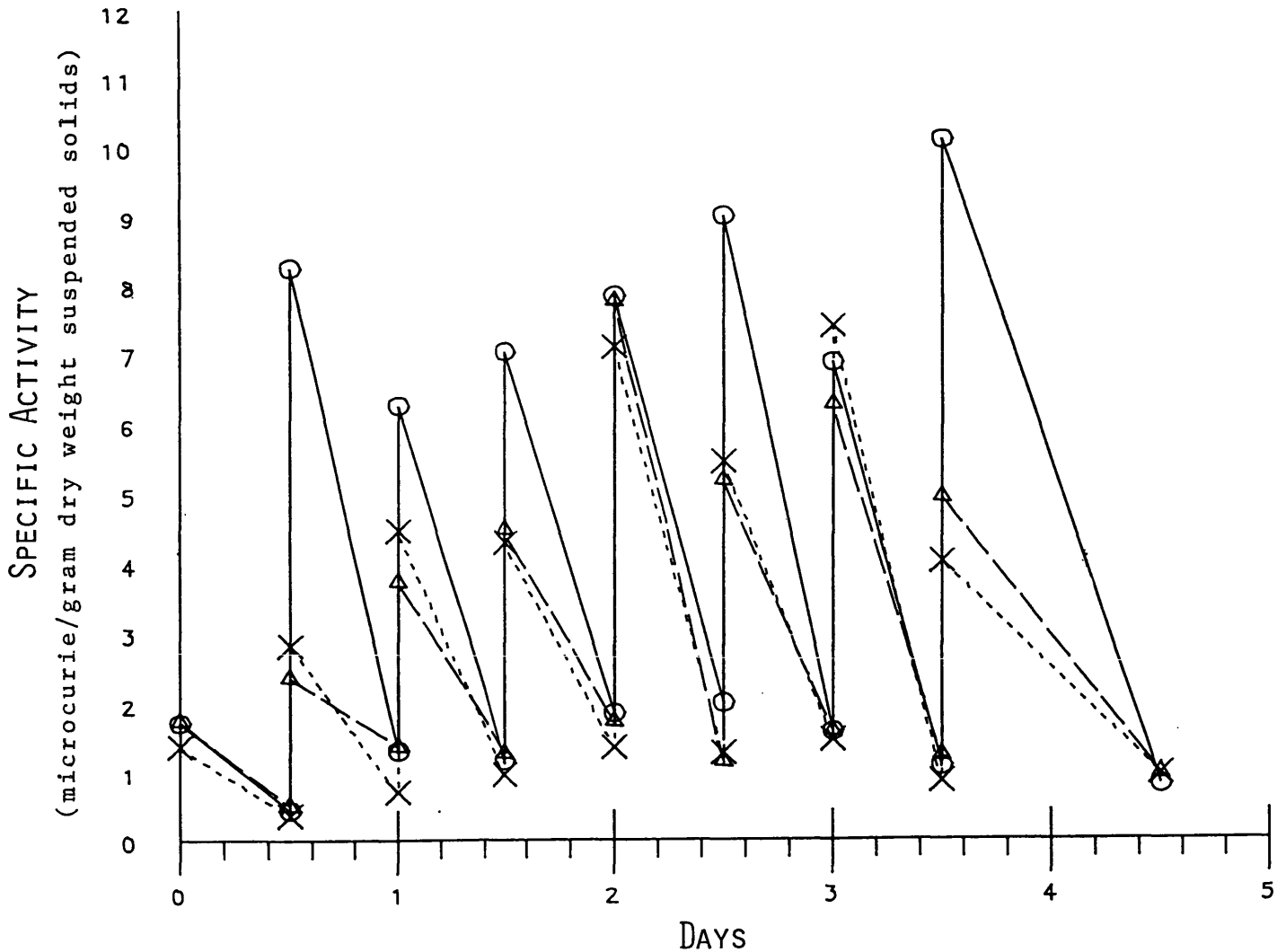

Figure 5. Time variation of the weight specific radioactivity of suspended solids in the aquaria (key as in Figure 3).

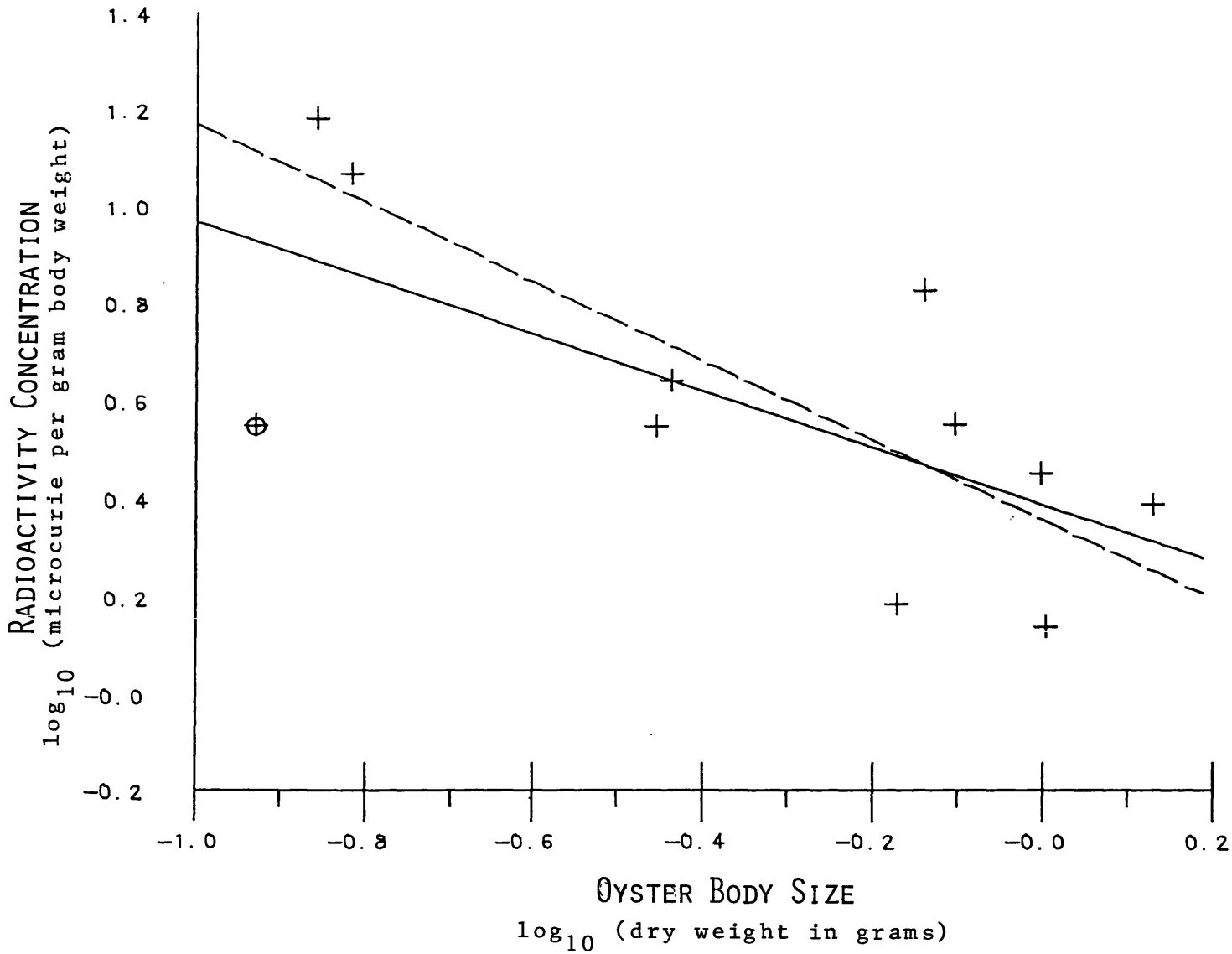

Figure 6(a). Variation in the weight specific uptake of radioactivity with oyster body size in aquarium No. 1. The linear regression line (_)was determined using a least square method after the logarithmic transform. The dotted line (--) was the regression line when the out-lier (⊕)was eliminated for the calculation. (T = 4.5 days)

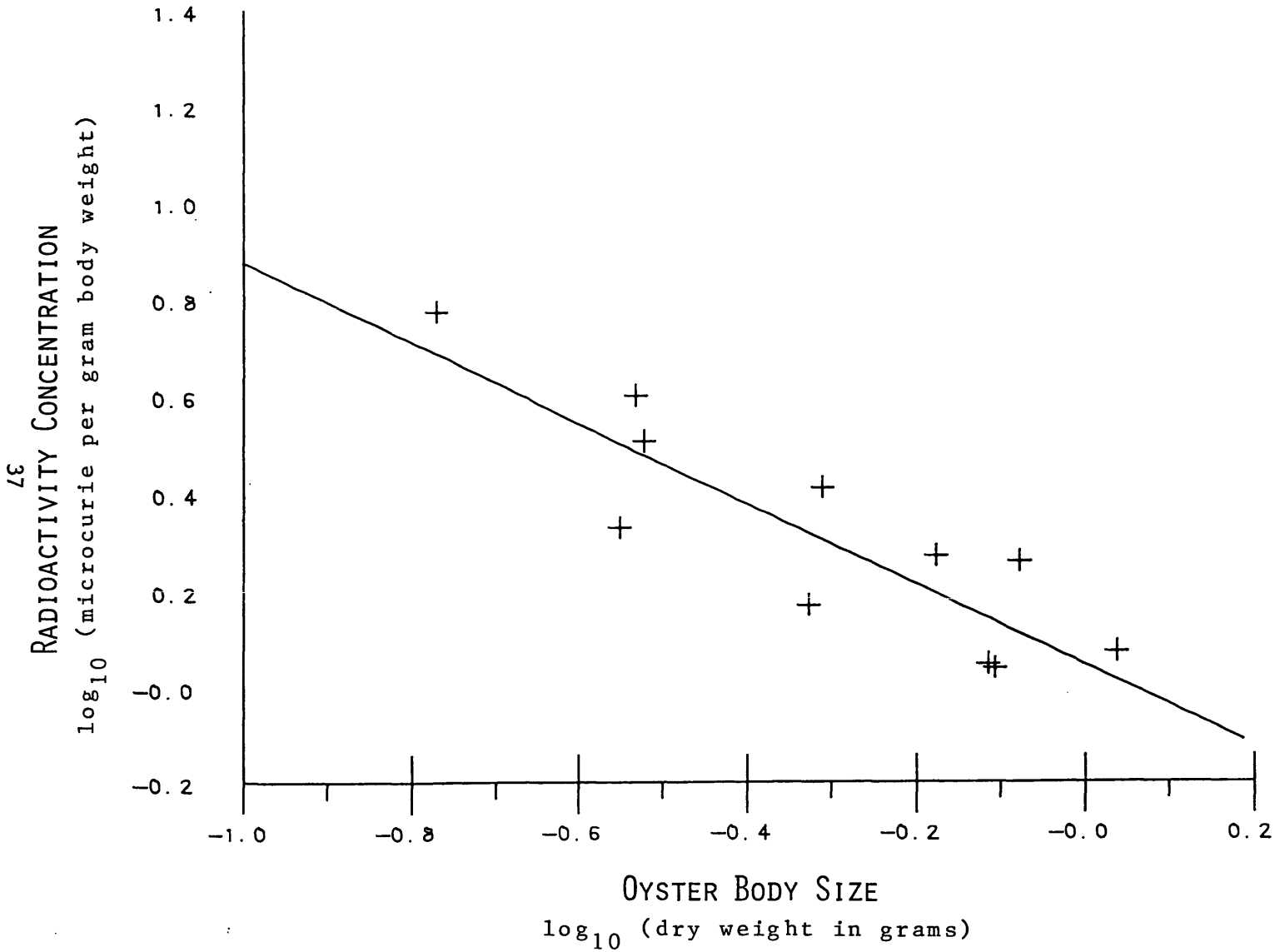

Figure 6(b). Variation in the weight specific uptake of radioactivity with oyster body size in aquarium No. 2. (T = 4.5 days)

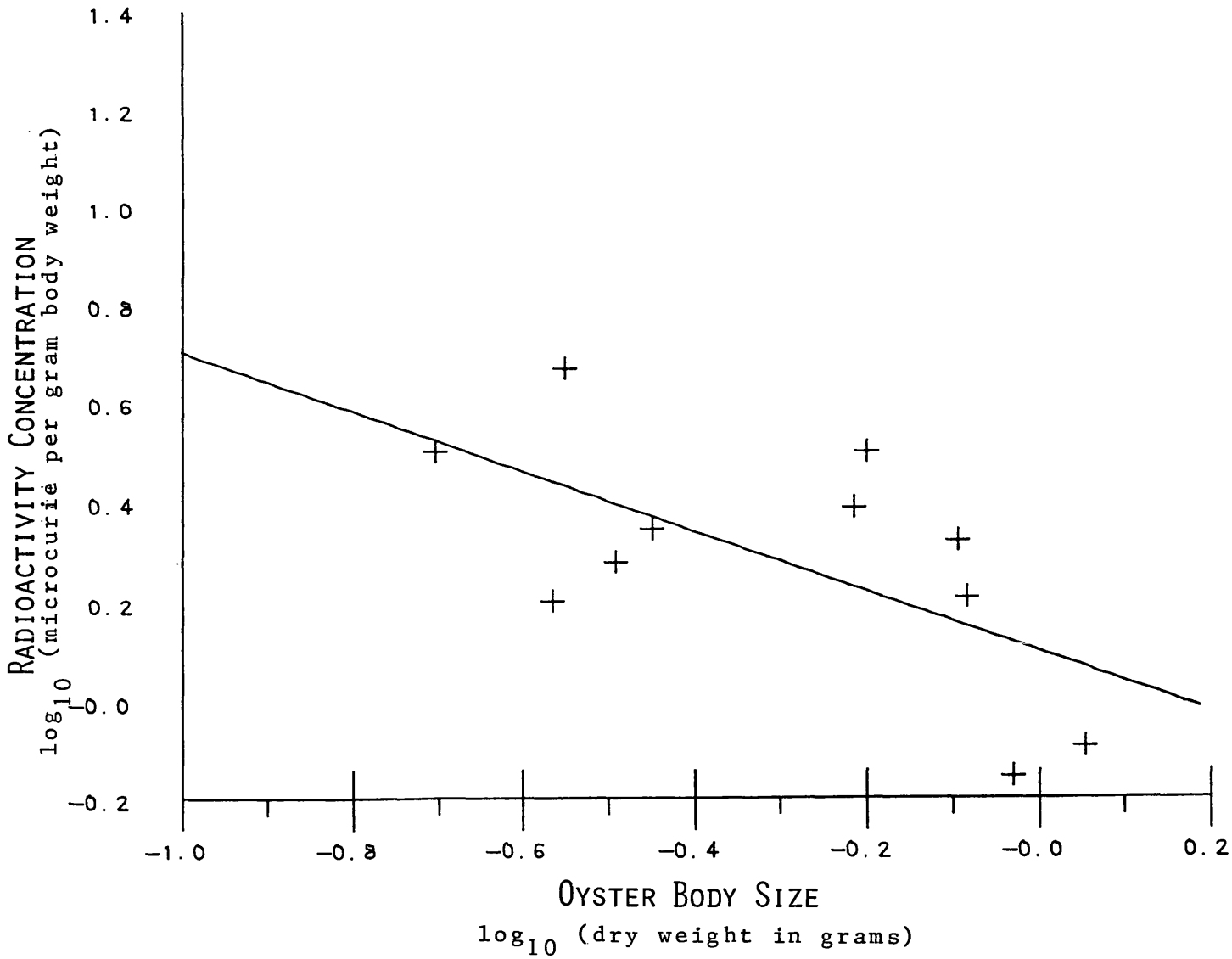

Figure 6(c). Variation in the weight specific uptake of radioactivity with oyster body size in aquarium No. 3 (T = 4.5 days).

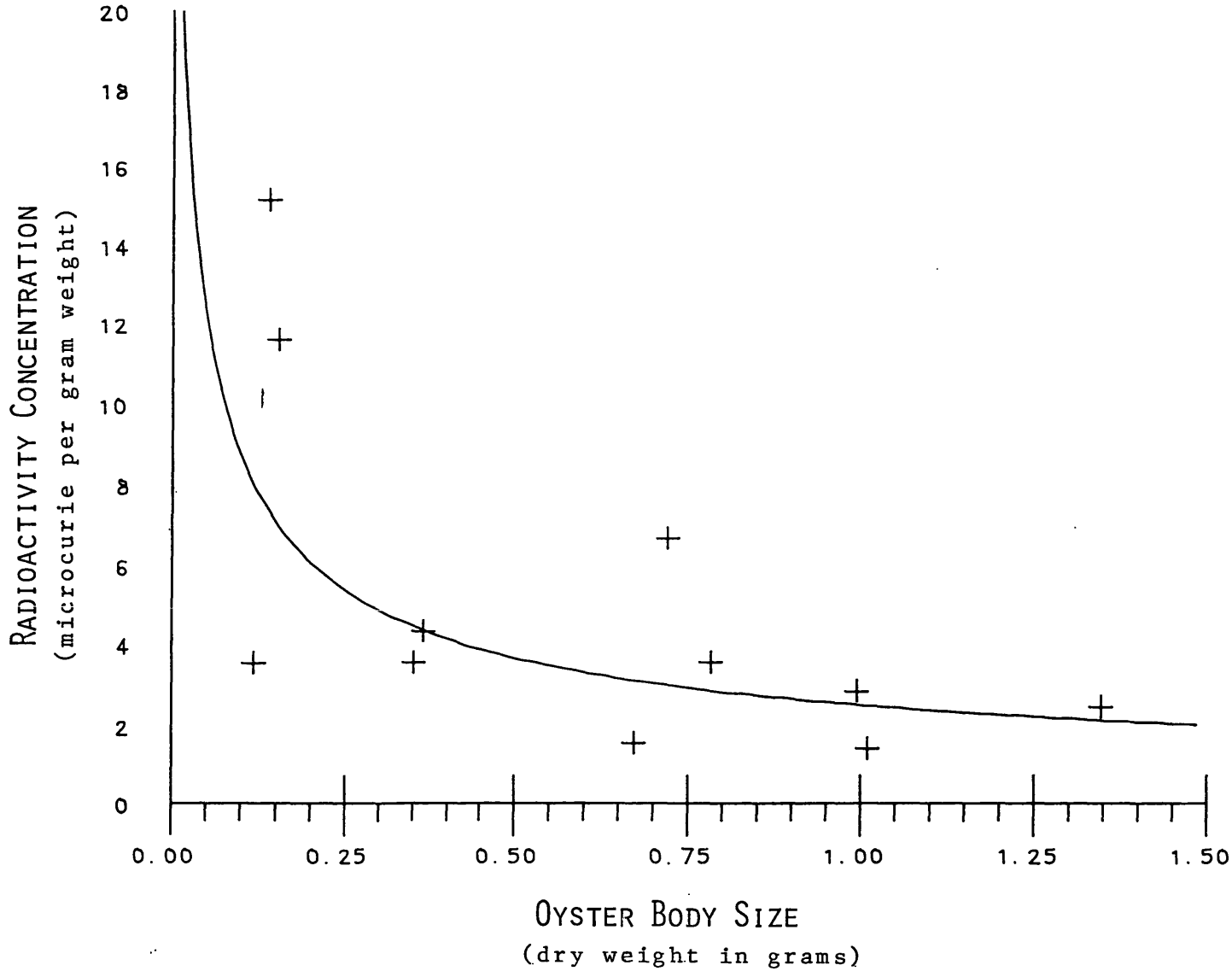

Figure 6(d). Variation in the weight specific uptake of radioactivity with oyster body size for aquarium No. 1. The actual measurements of the uptake rates and body sizes are shown. The curve is the back-transform of the figure in 6(a).

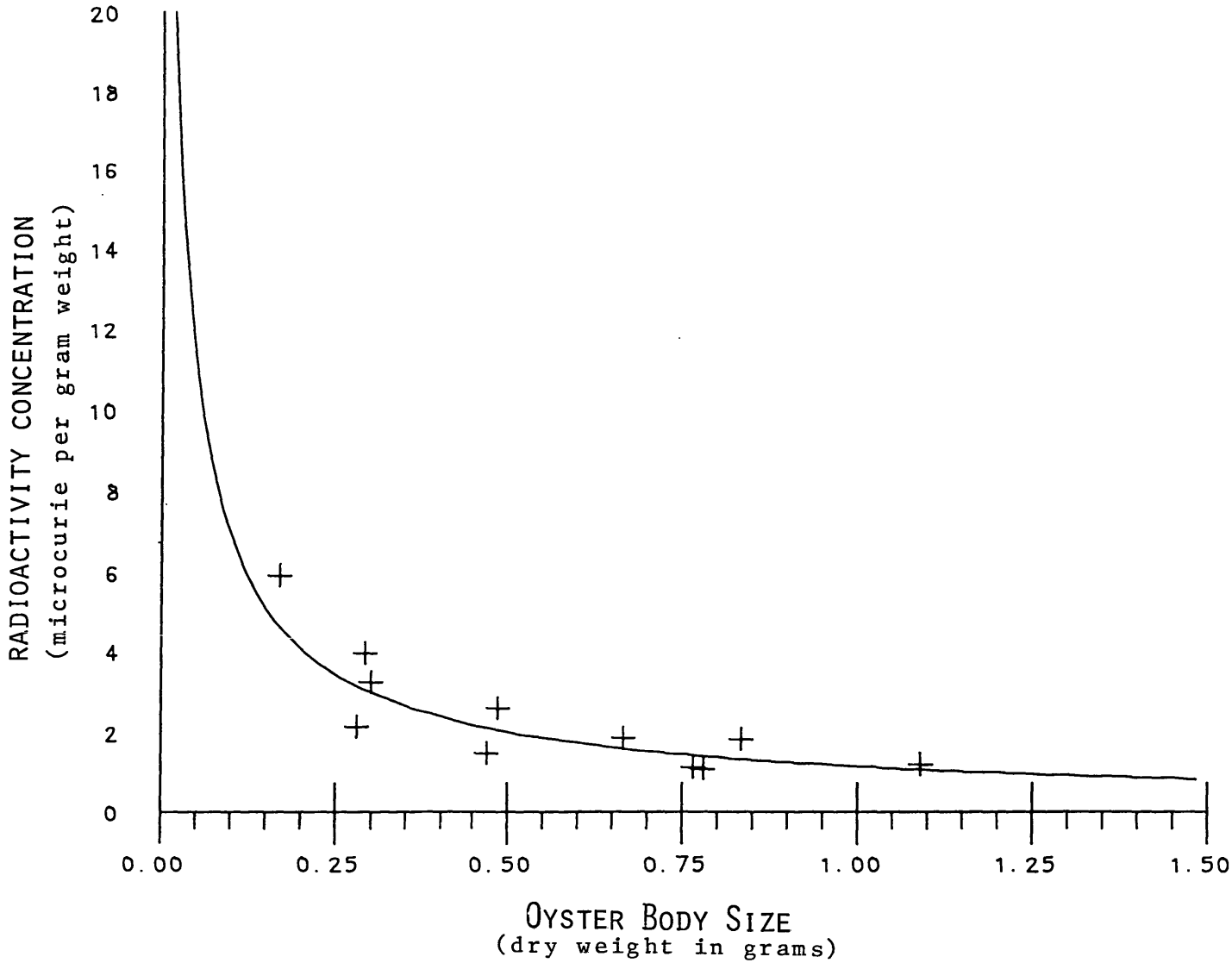

Figure 6(e). Variation in the weight specific uptake of radioactivity with oyster body size for aquarium No. 2.

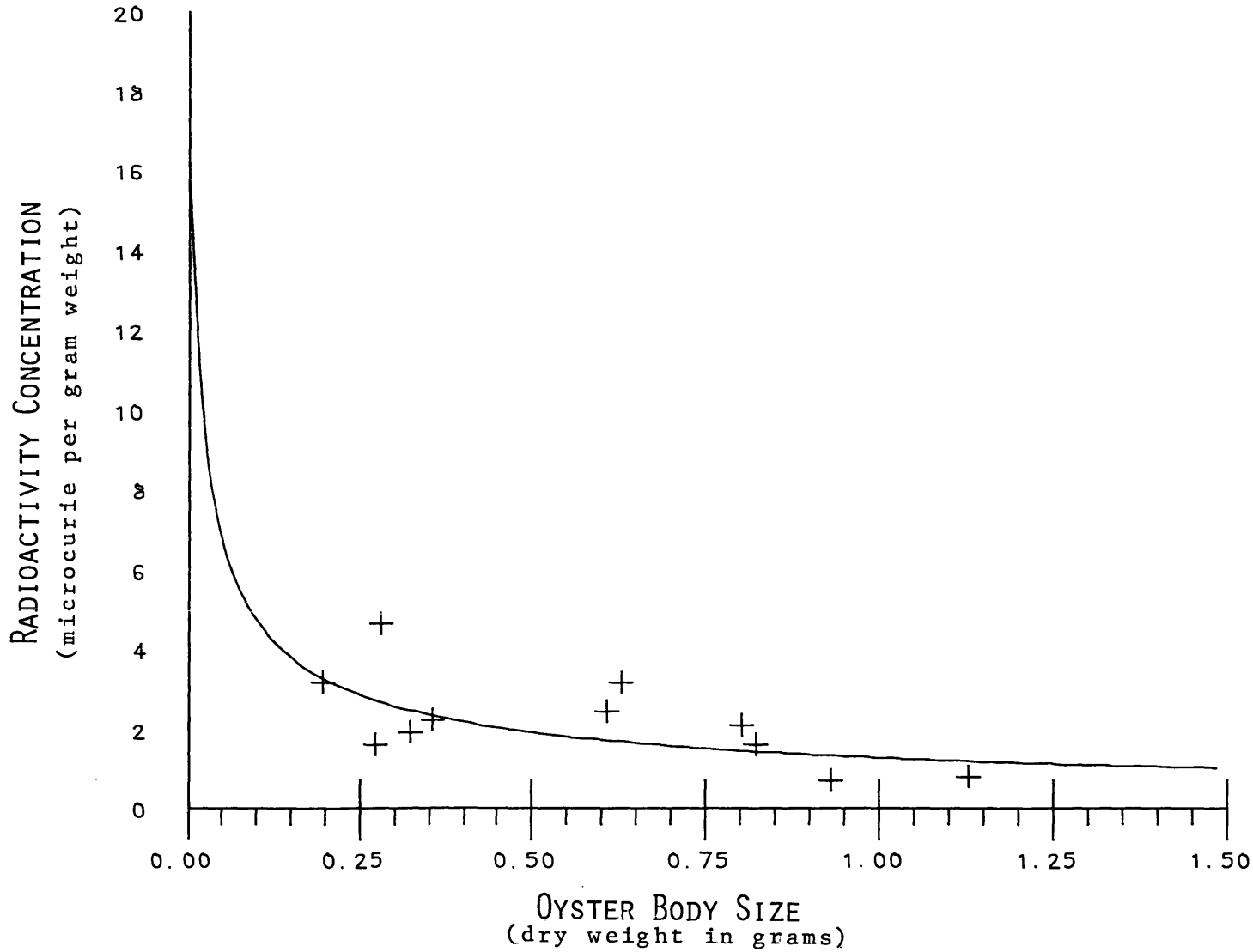

Figure 6(f). Variation in the weight specific uptake of radioactivity with oyster body size for aquarium No. 3.

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