Bruce E. Thompson

VARIATION IN BENTHIC ASSEMBLAGES

One of the goals of monitoring is to detect changes in biological assemblages caused by effluent discharge; however, these changes can only be interpreted with knowledge of natural fluctuations in existing assemblages. Sampling strategies, including the number of grab samples necessary to detect significant changes, have been debated often. This paper provides an analytical basis for such monitoring decisions.

The purposes of this study were: (1) to examine variation in the composition of benthic assemblages on the mainland shelf at different scales of sampling and various levels of contamination, and (2) to determine the appropriate number of replicate grab samples necessary in future sampling programs to detect changes in the assemblages.

Trends in species composition, numbers of species, individuals, and biomass along a generalized outfall gradient were similar at both the kilometer sampling scale and the meter sampling scale. There were, however, differences in the sample variation at the two scales; the smaller scale samples were always less variable.

Changes in the species composition of assemblages along outfall gradients can be demonstrated with single samples as well as with replicated grab samples. The number of replicate grab samples necessary to detect various levels of change in the numbers of species and individuals are presented, but usually are too numerous to be practicable.

METHODS

The region considered extends from Pt. Conception to the U. S. - Mexico Border along the mainland shelf (44-200m) of the southern California borderland (Figure 1). The data presented are based on samples collected during three different surveys: The 60-m Control Survey (Word and Mearns 1979), the Synoptic Survey (Bascom 1978), and the Replication Study (Word *et al.*, 1980) plus two new replicated sites not previously reported (Table 1).

Since this study was concerned with variation it was important to collect and analyze each grab sample in an identical manner. All samples were collected from soft sediment substrates using a 0.1 m^2 modified Van Veen grab and screened through a 1.0 mm sieve. The organisms from each sample were identified to the lowest taxon practicable. Consistent and standard taxonomic practices were used to ensure maximum comparability among the samples.

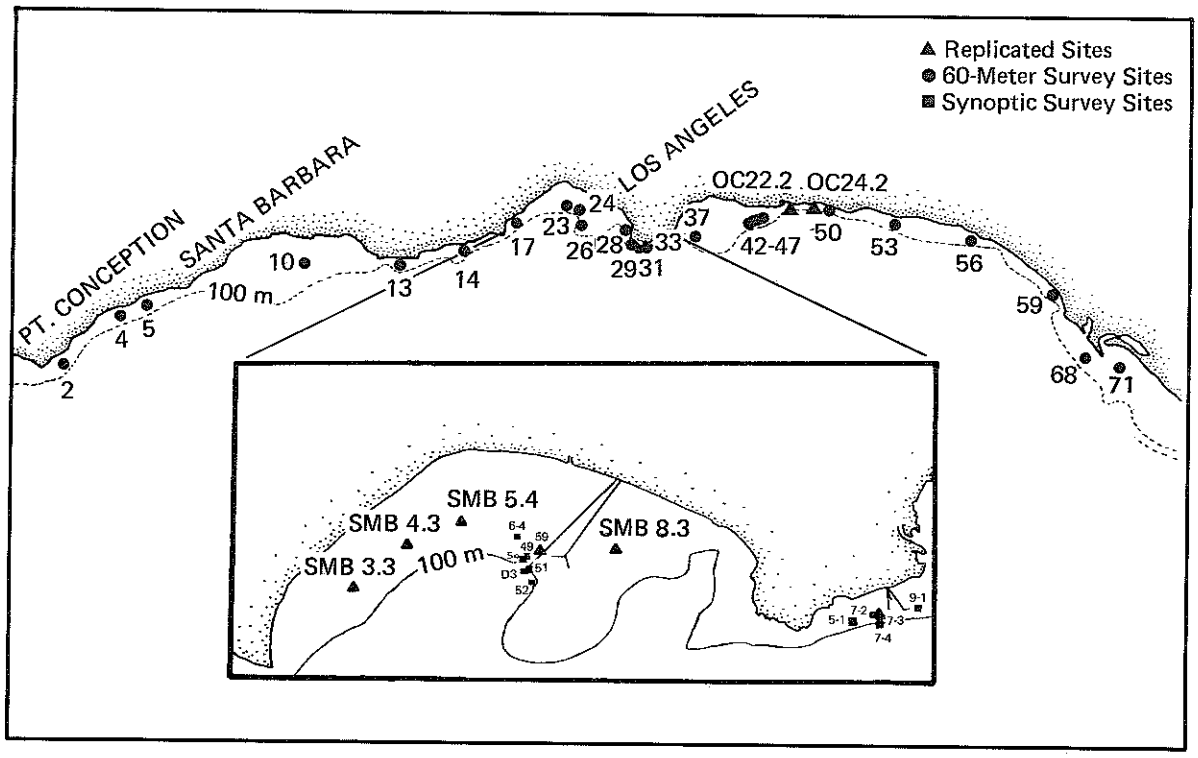

Figure 1. Chart of Study Area

Large scale variations (kilometer scale) within each zone were examined using the 60-m and Synoptic Survey data (Table 1). Thirty-six sites were selected for analysis (Figure 1). The selection of sites to be used was objective, but because of differences in the level of identification of the species in some samples, truly random site selection could not be made.

Small scale variations (meter scale) were examined using replicated grab data. Ten replicate grab samples from 7 sites and 9 replicates from one site (SMB 8-3) were collected (Table 1). All samples were from near 60 m, except OC 24-2 which was at 44 m. The replicate samples were all collected as follows: when the ship was on station, a marker buoy was dropped. The ship was then repositioned and the bow was kept on the buoy as each replicate sample was taken. We estimate that the replicates were collected within a 10-20 m diameter.

Ideally, when studying spatial variation, all samples should be collected at the same time or season to avoid temporal variation effects. However, along the coast of southern California, in water deeper than 30 m, large temporal changes do not occur. Fauchald and Jones (1978) reported only minor changes in community composition in a one year sampling sequence; these were mainly due to recruitment pulses of 1 or 2 species. Additionally, their results differ little from earlier studies by Hartman (AHF:USC 1959) and others (Jones 1969; Wintz and Fauchald 1972) demonstrating very stable assemblages (temporally) on the mainland shelf of the region. Therefore, in this paper, temporal changes in the composition of assemblages are assumed to be negligible.

STATISTICAL ANALYSES

It is helpful to begin by clarifying the differences between several parameters of biological assemblages. Estimates of total numbers of species, individuals, and biomass are commonly used

Table 1. Listing of study sties, 60 m and Synoptic Survey sites are listed by $\sum$ ranks. Other data from these sites may be obtained from references listed in text. *Indicates sites omitted from analysis of within and between zone variation, **indicates sites not previously reported.

	60 m and Synoptic Survey sites		Replicated Sites		
	Station	$\sum$ ranks	Station	Date	Depth (m)
CONTROL	13	5			
	71	6			
	59	9	OC 22.2	22 Jan 80	60
	4	19			
	5	26	OC 24.2	22 Jan 80	44
	68	26			
	53	28	SMB 3.3	9 May 79	58
	47*	28			
	14	29	SMB 4.3	9 May 79	61
	37*	30			
	50	30			
	SMB 6.4*	31			
	56	33			
TRANSITION	2	34			
	28	34			
	46	36	SMB 5.4	9 May 79	58
	42	38			
	17*	43	SMB 8.3	11 May 79	61
	10	47			
	45	48			
	23	49			
	26	54			
	29	54			
	24	60			
CONTAMINATED	PV 9.1	71			
	31*	75	SMB 59**	19 Nov 81	64
	PV 6.1	75			
	52	78	PV 7.3**	8 Feb 82	60
	PV 7.3	80			
	D3	81			
	33	83			
	PV 7.2	85			
	PV 7.4	85			
	50	93			
	51*	95			
	49	99			

to describe and compare benthic samples. It is conceivable, however, to have two collections with identical values of these parameters, but with entirely different species represented in each. Therefore, it is necessary to compare measures of which species are present as well as measures of how many species are present. In this study, measures of both types were used.

Variations in number of species, number of individuals, and biomass are expressed as coefficients of variation (C.V. = $\frac{\text{standard deviation}}{\text{mean}}$ x 100) so that samples may be compared directly. Variation in the species composition of each sample was evaluated by means of the Bray-Curtis similarity index (Smith 1976).

Precision (D), a measure of the accuracy of estimated mean values, is the ratio of the standard error to the mean. It was calculated as replicate samples were added; thus allowing an evaluation of replication effort required to obtain a desired level of precision (Elliott 1977).

The number of replicate grab samples required to detect 10, 20, and 30 percent changes in mean values of total species and total individuals was also calculated using the method of Sokal and Rohlf (1969).

The level of replication necessary to adequately detect changes in species composition was evaluated using Information Loss Analysis (Smith 1976). The maximum amount of "Information" (equal to similarity) is assumed for 10 replicates and is considered as zero information loss. As replicates are removed, the resulting similarity matrix is correlated to the matrix for 10 replicates and Information Loss is calculated as $(100)(1-r^2)$ where r is the product-moment correlation coefficient (Smith, 1976).

QUANTIFICATION OF THE OUTFALL GRADIENT

This paper assumes that a gradient of biological changes results from a gradient of outfall conditions. Measurements of three sediment parameters that reflect the gradient were made from grab samples collected concurrently with the samples used for the biological information (60-meter and Synoptic Survey data). Total volatile solids (TVS) was used as a measure or organic material, biological oxygen demand (BOD) as a measure of microbial respiration, and chromium (Cr) levels as a measure of general contamination. All were measured in the upper 2 cm. The primary source of chromium in sediments is from municipal outfalls (Jan and Hershelman, 1980). Methods of analysis for these three parameters are detailed in Word and Mearns (1979).

To demonstrate that the three sediment parameters are correlated and can be used together as outfall gradient indicators, Kendall's Coefficient of Concordance (W) (Siegel, 1956) was calculated. The value of $W = 0.86$ is significant at $\alpha = 0.5$ indicating a positive correlation between the rank orders of all three parameters. This demonstrates that the three parameters reflect the same gradient.

To establish a generalized gradient, first the values of each parameter (TVS, BOD, Cr) were ranked separately over all 36 sites; then the rank values of the three parameters at each site were summed. Arranged in numerical order, the sum of the ranks (Σranks) at each site will be used to represent the outfall gradient in the study area. The lowest value, 5, represents the normal or "Control" end of the gradient and the highest value, 99, represents the "Contaminated" end of the gradient. This generalized numerical gradient is distinct from a strict geographical gradient although in some areas, such as off Orange County, they may coincide.

BIOLOGICAL CHANGES ALONG THE GRADIENT

The total number of species, total number of individuals, and the total biomass (alcohol wet weight) at each of the 36 sites selected from the 60-meter and Synoptic Survey were plotted along the outfall gradient (Figure 2). These three biological parameters each demonstrated different levels of correlation to the gradient (Σranks); the number of species and total biomass were both significantly correlated to the gradient, but the total number of individuals was not.

To evaluate further how the outfall gradient influences organisms, the distributions of three species, the ophiuroid *Amphiodia urtica*, the pelecypod *Parvilucina tenuisculpta* and the polychaete *Capitella capitata*, along the gradient were examined (Figure 3). These three species have been reported previously as indicators of different levels of pollution (Word *et al.* 1980).

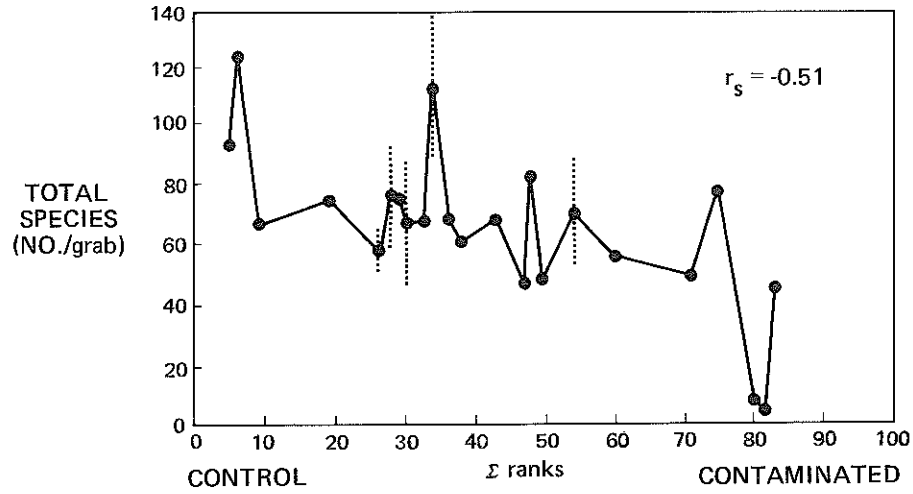

Figure 2a. Numbers of species in samples along generalized outfall gradient. Vertical broken lines represent range of values for sites with same Σ ranks.

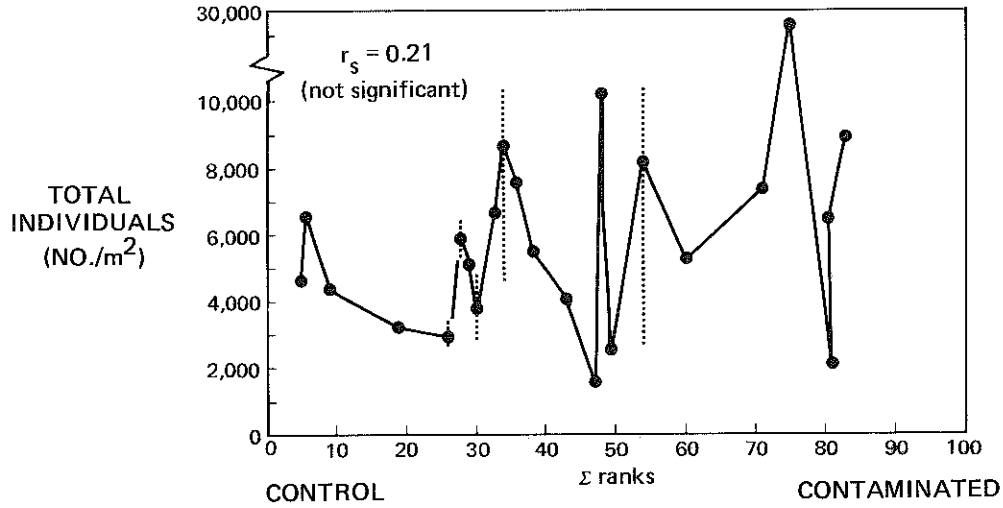

Figure 2b. Densities of grab collected invertebrates along generalized outfall gradient. Vertical broken lines represent range of values for sites with the same Σ ranks.

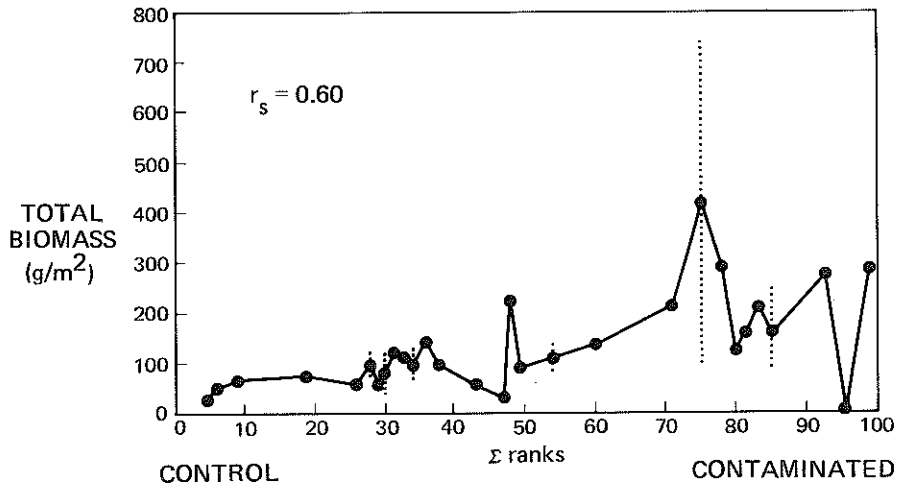

Figure 2c. Total Biomass (alcohol wet wt.) at sites along generalized outfall (gradient). Vertical broken lines represent range of values for sites with same Σ ranks.

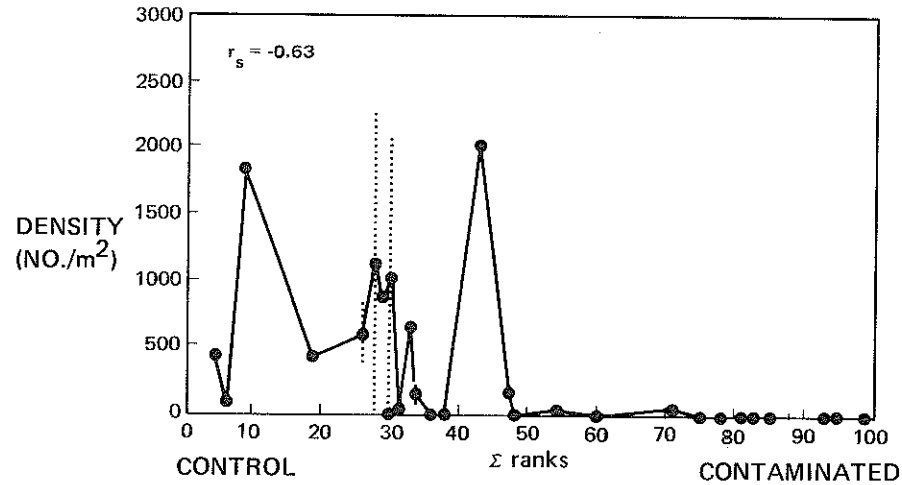

Figure 3a. Densities of *Amphiodia urtica* along generalized sewage outfall gradient. Vertical broken lines represent range of values for sites with same Σ ranks.

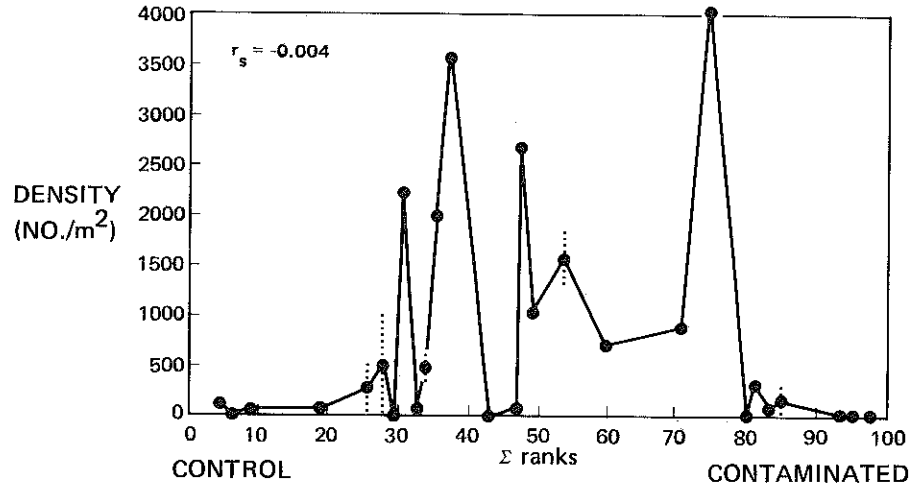

Figure 3b. Densities of *Parvilucina tenuisculpta* along generalized sewage outfall. Vertical broken lines represent rage of values for sites with same Σ ranks.

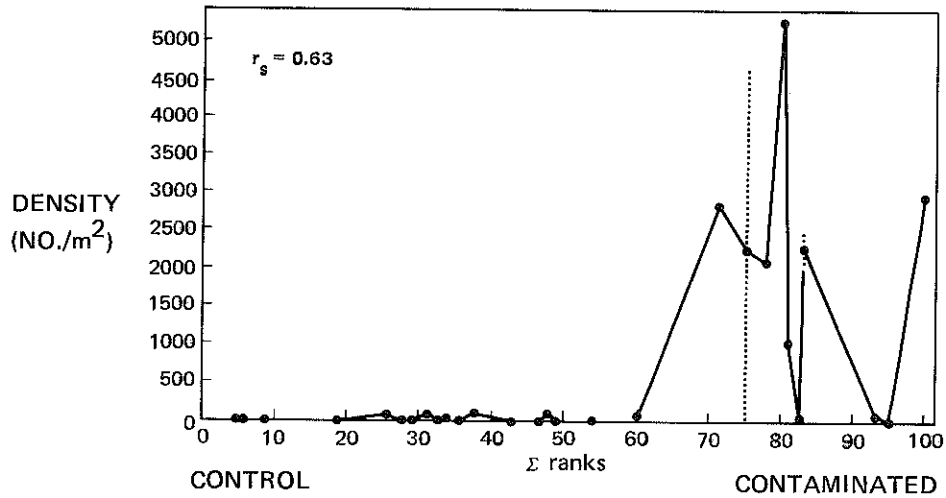

Figure 3c. Densities of *Capitella capitata* along generalized sewage outfall gradient. Vertical broken lines represent range of values for sites with same Σ ranks.

It is evident that *A. urtica* lives mostly in control areas and does not live in areas influenced by waste waters. This species was described as "the most abundant and wide-spread invertebrate on the mainland shelf of southern California" by Barnard and Ziesenhenne (1961). Jones (1969) reported *A. urtica* as the dominant species of assemblages from most natural areas on the mainland shelf. *P. tenuisculpta* occurs in highest densities in the middle of the gradient; it was not significantly correlated to the gradient, probably due to this non-linearity. The distribution and abundance of *P. tenuisculpta* in the southern California Borderland was recently described by Jones and Thompson (M.S.). While widespread throughout the Bight, it was found in highest densities near waste outfalls in Santa Monica Bay and off Palos Verdes. *C. capitata* occurs in highest densities in the immediate vicinity of outfalls and is only rarely collected in more normal areas. This species has been shown to occur near sewage outfalls and other polluted areas by many other workers (Reish 1959; Bellan 1967; Pearson 1972; Word *et al.* 1977).

DESIGNATION OF ZONES

The portions of the gradient where obvious increases and decreases in the abundance of *A. urtica, P. tenuisculpta,* and *C. capitata* occur demonstrate the existance of rather discrete zones (Figure 3). To facilitate further examination of changes in benthic assemblages along the gradient, boundaries between zones were established. It should be emphasized the boundaries selected do **not** delimit distinct zones *per se* because the gradient is a numerical one, not a geographical one. Rather, they aided in determining which of the 36 sites could be grouped as a sample of each zone so that average values for several biological parameters could be calculated and compared between zones.

The limits of the "Contaminated" zone were most easily determined. Between Σranks 60 and 70 there were obvious increases in abundance of *C. capitata* and total biomass. Thus, 65 was selected as the boundary between the "Contaminated" zone and the "Transition" zone.

The boundary between the "Control"and "Transition" zones was more difficult to designate. It appears that between Σranks 30 and 40 *A. urtica* begins to decrease and *P. tenuisculpta* begins to increase; there is some apparent overlap at the ends of their ranges. Thirty-five was subjectively selected as the boundary between the Control and Transition zones.

BENTHIC ASSEMBLAGES IN EACH ZONE

It is important to point out that within each zone some apparently inconsistent results were obtained. Several sites received the same Σranks, but differed considerably in their faunal composition. Just why sites with similar outfall gradient characteristics would have such different assemblages is not clear.

These four inconsistent sites and two sites where incomplete information was available were omitted from further analyses (see Table 1), leaving 30 sites, 10 in each zone.

Using the 10 sites in each zone as replicates, the average densities and coefficients of variations for the most abundant species in each zone were calculated. *A. urtica* was the most abundant organism in the Control zone, *P. tenuisculpta* was the most abundant organism in the Transition zone and *C. capitata* was the most abundant in the Contaminated zone.

The densities of these three species from the replicated sites demonstrated similar patterns. *A. urtica* was most abundant at stations OC 22-2, OC 24-2, SMB 3-3, SMB 4-3; therefore, these sites are considered to be control stations. *P. tenuisculpta* was most abundant at SMB 5-4, but at SMB 8-3 the polychaete *Myriochele* sp. was most abundant and apparently represents a large

patch of these animals; these two sites are considered to represent Transition zones. *C. capitata* was most abundant at PV 7-3 and it is, therefore, considered a Contaminated site. Station SMB 59 also represents a Contaminated site. Although *P. tenuisculpta* was the most abundant species there (more characteristic of Transition sites) a considerable number of *C. capitata* were also collected.

A comparison of the variation in the densities between large scale (km) sampling and replicate grab (m) sampling for *Amphiodia urtica, Parvilucina tenuisculpta* and *Capitella capitata* is shown on Table 2. Variation was always less within small scale samples than in larger scale samples.

Comparison of the variation in number of species, individuals, and biomass in each zone are shown in Table 3. Variation in these parameters was generally lower than variation in individual species abundances (Table 2). Variation nearly always exceeded 30% at the km scale and was usually lower among the replicates.

Table 2. Comparison of mean abundances (#/grab) and coefficients of variation for three zone indicator species sampled at two spatial scales. Km scale data from 60-m and Synoptic Survey data (n = 10); m scale date from Replicated grab data (n = 10).

	Zone =	Control		Transition		Contaminated	
Species	Scale =	km	m	km	m	km	m
Amphiodia urtica	$\bar{X}$	105.0	140.4	5.2	4.9	0	0
	C.V.	74.3	23.0	213.9	57.7	0	0
Parvilucina tenuisculpta	$\bar{X}$	8.4	4.3	140.4	201.9	24.5	93.5
	C.V.	169.0	57.6	77.6	31.6	142.5	49.6
Capitella capitata	$\bar{X}$	0.2	0.5	1.3	1.2	245.9	101.9
	C.V.	316.2	79.1	149.7	117.1	73.0	36.8

Table 3. Comparison of means (#/grab) and coefficients of variation of three biological parameters sampled at two spatial scales. Km scale data from 60-m and Synoptic Survey data (n = 10, except in contaminated zone where n = 5); m scale data from replicated grab data (n = 10).

	Zone =	Control		Transition		Contaminated	
Parameter	Scale =	km	m	km	m	km	m
Total Species	$\bar{X}$	75.1	74.5	72.6	122.3	40.4	40.7
	C.V.	27.0	14.5	35.0	8.7	68.6	19.2
Total Individ.	$\bar{X}$	480.3	455.9	658.0	1513.6	1110.0	425.5
	C.V.	30.0	21.4	64.6	15.0	100.7	21.4
Total Biomass	$\bar{X}$	7.3	9.6	10.5	10.9	22.3	22.7
	C.V.	42.4	42.5	48.5	51.4	79.2	40.0

DIFFERENCES BETWEEN ZONES

The most abundant species in each zone was the same at both sampling scales (Table 2). Variation was lowest in the zone in which each was most abundant. Using the 60-m and Synoptic

Survey data, other differences in species composition between the three zones may be shown most clearly using a 2-way table of abundances (Table 4). In general, there are large differences in species abundances between zones. A few species, such as *Spiophanes missionensis*, were equally abundant in two of the zones.

Using the replicate data, similarity indices within the replicates were always significantly lower than similarities between the zones (Dyer's test; see Smith 1982) further demonstrating a strong gradient of species composition.

Trends in total species, individuals, and biomass between zones were usually consistent at both scales sampled. Numbers of species were highest in the Transition zone and lowest in the Con-

Table 4. Two-way table of average abundances of 10 most abundant species in each zone.

	Control	Transition	Contaminated
		(X/m^2)	
Control			
1. *Amphiodia urtica*	1050	520	0
2. *Spiophanes missionensis*	581	542	15
3. *Cistena californiensis*	422	196	58
4. *Phoronis sp.*	203	132	0
5. *Maldane sarsi*	45	1	0
Transition			
1. *Parvilucina tenuisculpta*	84	1404	245
2. *Euphilomedes carcharodonta*	86	560	5
3. *Myriochele sp. m*	2	472	0
4. *Prionospio steenstrupi*	32	287	38
5. *Axinopsida serricata*	123	207	35
6. *Chloeia pinnata*	41	187	31
7. *Euphilomedes producta*	64	112	5
8. *Mediomastus sp.*	3	102	100
9. *Spiochaetopterus costarum*	45	47	24
10. *Heterophoxus oculatus*	47	48	1
Contaminated			
1. *Capitella capitata*	2	11	2459
2. *Mysella pedroana*	30	35	615
3. *Tharyx sp.*	29	37	213
4. *Schistomeringos sp.*	0	0	175
5. *Dorvillidae, U. I.*	0	0	186
6. *Oligochaeta*	0	0	63
7. *Notomastus sp.*	2	15	110
8. *Nereis procera*	1	0	68

were highest in the Transition zone and lowest in the Contaminated zone. No clear trend was found in numbers of individuals. Biomass increased steadily from Control to Contaminated sites.

Samples from the Contaminated zone usually had the most variation in all three parameters at both sampling scales. Samples from the Transition zone were least variable at the m scale, but samples from the Control zone were least variable at the km scale.

EVALUATION OF REPLICATION

The precision (D) of estimates of numbers of species and individuals was calculated as replicate samples were added at each site.

D for one sample was back-calculated; it indicated values within 20% of the mean at all sites except PV 7-3 (D = 20.2). The most precision was gained with the second replicate; additional replicates increased precision only slightly (Table 5). D values were always within 10% of the mean when all 10 replicates were considered. Twenty percent is considered to be an acceptable level of precision by most workers (Elliott 1977; Gonor and Kemp 1978).

The number of replicate samples necessary to detect various levels of change in numbers of species and individuals at each replicated site, with 80% probability and 95% confidence, were calculated and used to construct the curves in Figure 4.

Using the coefficients of variation listed in Tables 2 and 3, the number of replicates needed to detect a desired level of change may be determined. For example, the CV for numbers of species from the replicated sites in the Control zone was 14.5% (Table 3). To detect a 10% change (80% probability with 95% confidence), 34 replicate samples should be collected. Overall, to detect a 10% change in either species or individuals, the number of replicates is always more than could be sampled realistically. To detect a 30% change the number of replicates required are more reasonable and vary from 2 to 17.

Table 5. Precision, D, of estimates of total species and individuals at three levels of replication; D_1 = one sample, D_2 = two replicates, D_{10} = 10 replicates. (SMB 8.3 = 9 replicates). D values are expressed as percent error of the mean.

	CONTROL				TRANSITION		CONTAMINATED	
	OC 22.2	OC 24.2	SMB 3.3	SMB 4.3	SMB 5.4	SMB 8.3	SMB 59	PV 7.3
Total Species								
D_1	19.1	11.7	16.0	11.2	5.7	11.6	18.1	20.2
D_2	11.1	6.4	9.4	6.2	3.1	6.8	10.7	12.1
D_{10}	6.0	3.7	5.1	3.6	1.8	3.9	5.7	6.4
Total Indiv.								
D_1	27.1	14.1	24.2	20.0	14.8	15.2	14.1	28.7
D_2	14.7	7.6	13.7	12.0	8.1	9.1	8.2	16.5
D_{10}	8.6	4.4	7.6	6.3	4.7	5.1	4.5	9.1

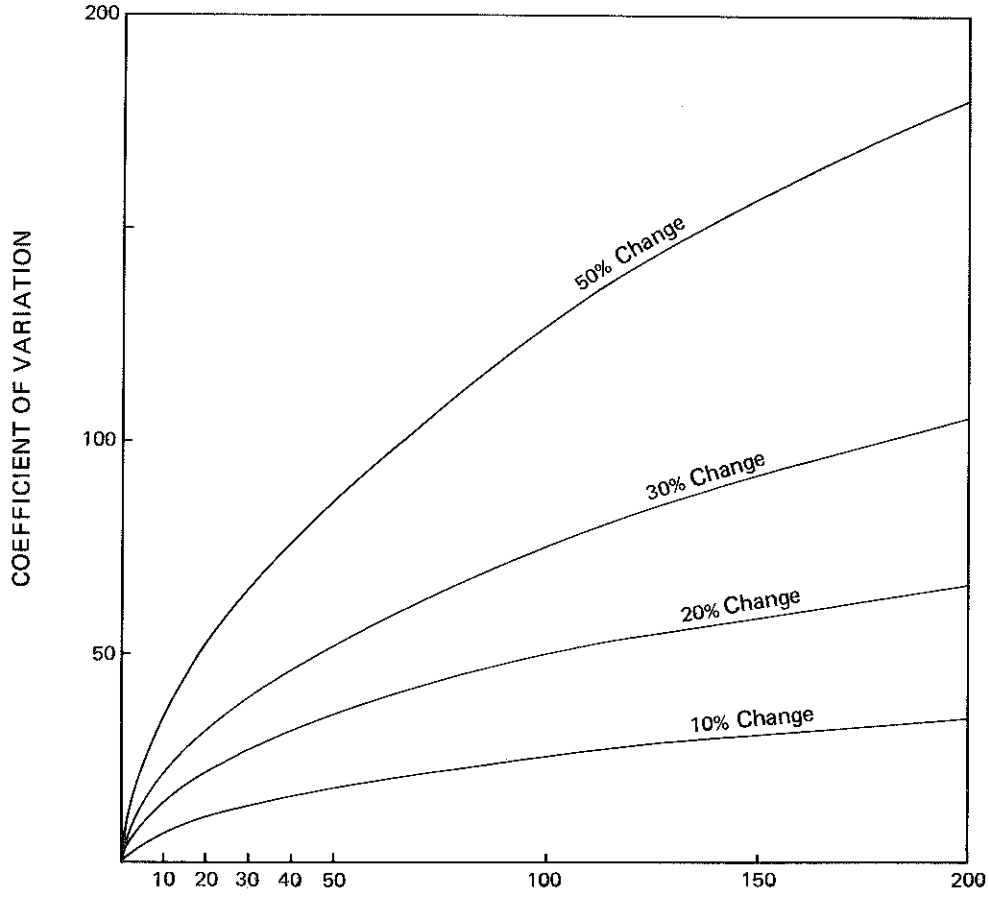

Figure 4. Graph of number of replicates necessary to detect 4 levels of change at a site with a probability of detection of 0.80 with 95% confidence.

Changes in numbers of species and individuals from Control to Transition to Contaminated areas usually range from 40% to more than 200%; thus, even fewer replicates, usually two, would be necessary to detect such changes. However, more conservative levels of change should be adopted to detect trends before assemblages completely change from representing one zone to another.

One must also consider which species are collected in each sample when determining levels of replication. Information Loss analysis indicated that similarities **between** sites can be determined equally well whether 1 or 9 replicates were used (Figure 5). This implies that differences in the species composition between zones may be demonstrated adequately using a single sample. Similar conclusions were reported by Cuff and Coleman (1979) using a somewhat different approach.

SUMMARY AND CONCLUSIONS

A generalized outfall gradient was quantified and divided into 3 zones: Control, Transition,and Contaminated. The most abundant species in each zone were usually distinct and they distinguished zones better than differences in numbers of species and individuals.

Variation in numbers of species and individuals was highest in the Contaminated zone samples. In all zones, the meter scale samples were less variable than the kilometer scale samples.

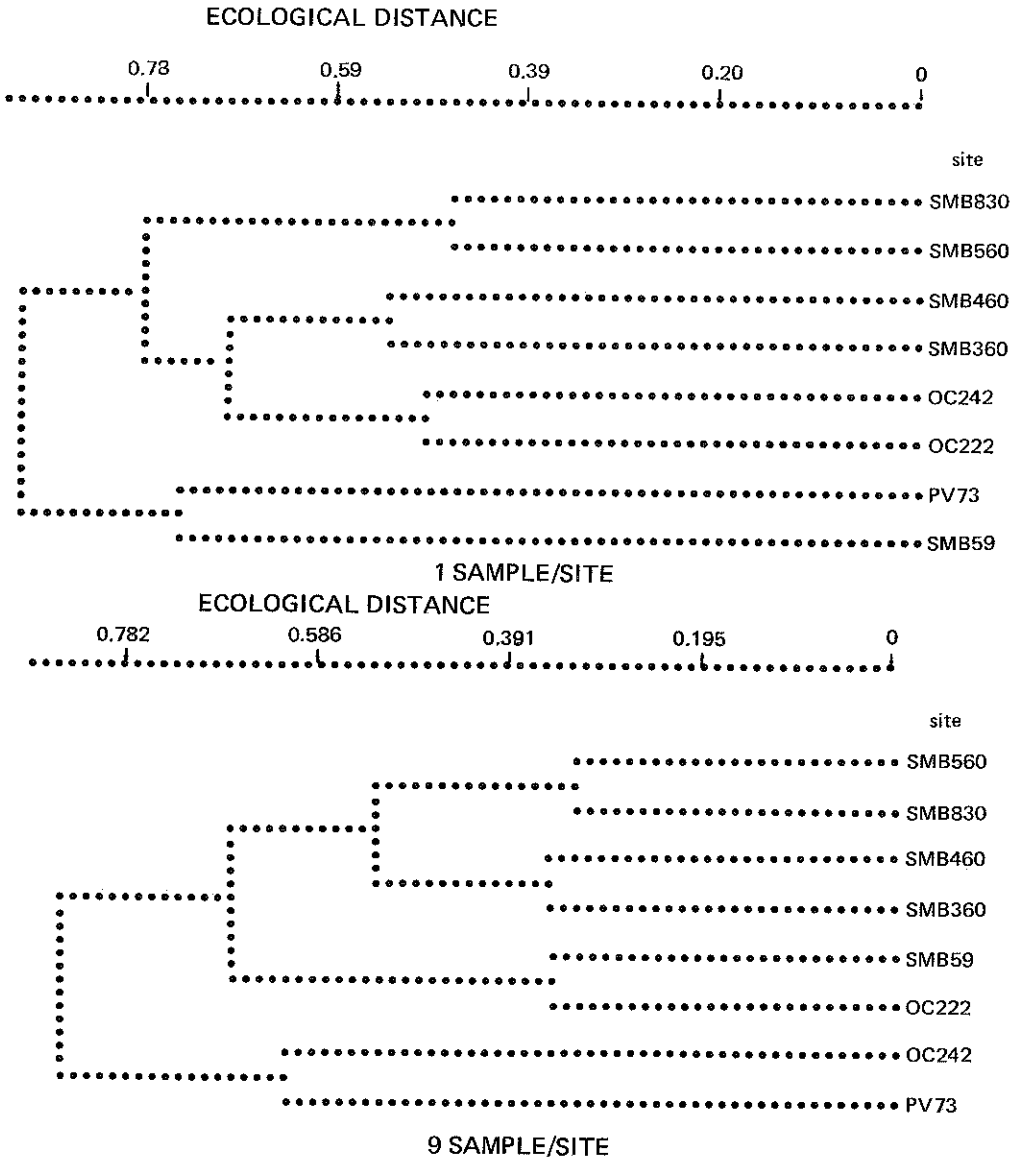

Figure 5. Dendrograms from Information Loss Analysis (Smith 1976). Ecological Distance = (similarity -1).

The similar trends in numbers of species, numbers of individuals, biomass, and species composition at both scales provide good evidence for the validity of these trends in this region.

Within the replicate samples, estimates of numbers of species and individuals were within 20% of the mean with 2 replicates at all sites.

With the information presented herein, future monitoring and survey programs can be designed more efficiently. Sampling efforts should be planned to ensure the maximum probability of detecting changes within time and money constraints.

The number of replicates necessary to detect changes can be determined from Figure 4 if a variance estimate is known.

Based on the results of this study, dichotomous sampling programs are suggested. Large areas can be covered by single samples to monitor for changes in species composition. Statistical comparison of single grab samples with the parameters reported herein for each zone can be made using the method presented in Sokal and Rohlf (1969). Additionally, at selected sites, 3-4 replicate samples should be collected to monitor changes in other assemblage parameters.

ACKNOWLEDGEMENTS

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