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Survey of Biological Effects of Toxicants upon Puget Sound Biota

IV. Interrelationships of Infauna, Sediment Bioassay and Sediment Chemistry Data

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NOTICE

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PREFACE

This report is the last in a series that describes some of the toxicological conditions of Puget Sound sediments. The first three reports described the results of surveys in which the toxicity of sediment samples was tested in laboratory bioassays. Distinct geographic trends were observed in the bioassay results. In initial qualitative examinations of available data, it appeared that those areas of the Sound in which toxicity was greatest were among the most contaminated. Also, the resident infaunal communities appeared to be changed in these same areas. The study reported here was initiated to examine more thoroughly (qualitatively) the interrelationships of the geographic trends in resident infauna, sediment bioassay and sediment chemistry data sets. A goal of the study was to determine if the three data types could comprise a "triad" of observations usable in a sediment quality index. A second goal was to determine values for the Index of Benthic Degradation based upon the benthic infaunal data. The Contracting Officer's Technical Representative for this study was Edward R. Long.

EXECUTIVE SUMMARY

Intensive environmental studies have been undertaken in Puget Sound by many groups, including NOAA initially under the auspices of the Marine EcoSystems Analysis (MESA) Puget Sound Project, and, later, under the Ocean Assessments Division (OAD). Bulk sediment analyses, sediment bioassays and histopathological studies have determined that some areas of Puget Sound are both highly contaminated and toxic to biota. However, the ecological significance of these findings remains to be determined. Although preliminary surveys had been conducted by NOAA through the National Marine Fisheries Service (NMFS) to collect and partially identify resident benthic infaunal communities, complete data on benthic infauna residing in contaminated and uncontaminated areas of Puget Sound were not available. The present study was initiated in a preliminary attempt to provide this missing information by first conducting species-level taxonomic identifications on previously-collected NOAA/NMFS samples. The data were then used to calculate the Index of Benthic Degradation (IBD) as developed for NOAA (O'Connor, unpublished report). As a further step in determining ecological significance, the data were correlated with concurrent sediment chemistry and sediment bioassay data (the "Triad" Index) to determine areas of significant environmental degradation in Puget Sound.

Archived benthic samples collected by NMFS from three urban embayments (Elliott Bay and the Duwamish, Commencement Bay Waterways, Sinclair Inlet) and two rural embayments (Case Inlet, Samish Bay) representing 12 stations were chosen for taxonomic analysis based on similarities in depth and sediment texture characteristics. Taxonomic data for these stations were combined with other benthic data that became available within the time frame of this study. These additional data included seven stations in Commencement Bay (Waterways) and four stations in Elliott Bay having similar depth and sediment texture characteristics to the 12 NMFS stations. Although these data sets were not completely compatible, useful information was obtained from combined and comparative analyses.

The complete data set for these 23 stations was examined for differences in taxon distributions between areas. Benthic communities in urban embayments were characterized by a high proportion of polychaetes and molluscs, a low proportion of arthropods and echinoderms and the virtual absence of sensitive amphipod families. The reverse situation existed in the rural embayments.

The complete benthic data set was used to calculate the IBD for urban versus reference areas. The results indicated that all the urban samples examined were significantly degraded.

Multi-variate statistical analyses of the benthic community data indicated that samples from the urban embayments were separable from those from the reference embayments, and that the Case Inlet reference area was more similar to the urban embayments than to Samish Bay. Samish Bay therefore appeared to be the least degraded of the two reference areas. Indications of differences between the urban embayments were substantiated by multi-variate analyses of combined data on sediment chemistry and benthic community structure. Inclusion of chemistry data with benthic data revealed substantial differences among the data for Elliott Bay, Sinclair Inlet and Commencement Bay Waterways, a pattern which was only suggested by analyses of benthic community data alone. The latter statistical analyses were restricted to the 12 NMFS samples because of

incompatible chemistry data for other stations. This resulted in a ranking of Commencement Bay Waterways, Elliott Bay and Sinclair Inlet, respectively, as the most to least degraded/pollution-affected based on the limited samples examined. Case Inlet was more similar to Sinclair Inlet than to Samish Bay in the statistical test discriminating parameters.

Graphical comparisons of sediment bioassay data and sediment chemistry with benthic community structure, (the "Triad" Index) confirmed the results of the above analyses. Highest sediment levels of organic and metal contaminants generally corresponded with increased toxicity and changes in benthic community structure. The results of the Triad provide the most definitive evidence for a linkage between sediment contaminant levels and biological effects. All three components of the Triad are essential for comprehensive marine pollution assessment: chemical analyses alone provide no information on availability and toxicity to resident biota; community structure may be affected by factors other than contaminant effects; and, bioassays provide direct evidence of adverse contaminant effects under laboratory conditions.

The results of the present study provided information on relative environmental degradation/pollution in different areas of Puget Sound and illustrated the value of the Triad approach for comprehensive marine pollution assessment. The IBD was a useful method for comparing areas of degradation based solely on benthic community structure; as such, it could ultimately form part of the Triad Index. Recommendations are provided regarding testing and further development of these two Indices.

1.0 INTRODUCTION

This report documents the results of a study commissioned by NOAA (initially through the Marine EcoSystems Analysis (MESA) Puget Sound Project and, later, through the Ocean Assessments Division (OAD)) to determine benthic infaunal species composition in selected areas of Puget Sound for which sediment chemistry and sediment bioassay data were available. Species-specific infaunal taxonomic data had not previously been available over a wide range of contaminated and non-contaminated areas of Puget Sound. The infaunal species data determined for selected NOAA study sites were then combined with selected similar and reasonably compatible data from two other studies in Puget Sound (Municipality of Metropolitan Seattle (Metro) - Comiskey et al., 1983; U.S. EPA, Newport, Oregon - Swartz, unpublished data) in an attempt to determine relationships between sediment infauna, sediment bioassays and sediment chemistry in Puget Sound. Available data were used retrospectively to test relationships between measures, despite indications that there were problems with the compatibility of the data sets.

Previous studies have demonstrated the presence of high environmental levels of chemicals in sediments from different areas of the Sound (Malins et al., 1980, 1982), and that a variety of biological effects occur in areas with high levels of contaminants (Long, 1983; Chapman et al., 1983). In addition, direct evidence of toxicity from Puget Sound sediments has recently been provided (Chapman et al., 1982; 1983; 1984; in press; Swartz et al., 1982). However, although it is apparent that sediment samples from some areas of Puget Sound are chemically contaminated and toxic under laboratory conditions, these test conditions may not be reflective of real-world or in situ effects on resident organisms.

At present the chemical(s) responsible for the observed toxicities are not known, and the ecological significance of toxic chemicals to sediment-associated biota is unknown. Therefore, the intent of the present study was to provide initial data to confirm or reject the hypothesis that elevated chemical levels and/or positive toxicity test results indicate adverse effects on biota living in areas with these characteristics.

Taxonomic analyses were conducted on archived samples from 12 NOAA stations to determine what resident infauna were present at stations in Puget Sound representing a gradient of chemical contamination and toxicity. Stations were chosen based on the similarity of depth and sediment textures. Additional compatible data from four stations analyzed by Metro Seattle as part of their Toxicant Pre-treatment Planning Study (TPPS), and seven stations analyzed by the U.S. EPA (Newport, Oregon, Laboratories), were included and the combined infaunal data were analyzed by the Index of Benthic Degradation (IBD) recently proposed by NOAA for defining coastal pollution-induced degradation (O'Connor, unpublished report). Multi-variate statistical and other analyses were then applied to the combined toxicity, chemistry and infaunal data (from more limited numbers of stations than used for the IBD) to determine regional differences for comparison with the IBD results. The results of these analyses were used to identify areas of significant degradation in the Puget Sound ecosystem.

1.1 Objectives

The specific objectives of this study were:

1. To conduct species-level taxonomic identifications on archived NMFS benthic samples from Puget Sound.

2. To test NOAA's Index of Benthic Degradation (IBD) (O'Connor, unpublished report), using available benthic infaunal data from Puget Sound.

3. To determine interrelationships between resident infauna, sediment chemistry and sediment toxicity data, including any differences in the benthic infauna at sites with different degrees of chemical contamination and sediment toxicity.

2.0 METHODS

Sediment texture and depth can greatly influence benthic infaunal community composition (c.f. Comiskey et al., 1983), and the protocol for the IBD requires comparisons of stations with equivalent sediment textures and depth. Accordingly, stations chosen for analysis represented the narrowest possible range of variation for the available data sets: mean grain size (Phi), 5.8-8.5; sand:mud ratio, 0.02-0.23; depth (m), 7-60. The only exception (sediment texture) was Station 11 in Samish Bay (Fig. 1), for which taxonomic analyses were conducted prior to sediment analyses.

Stations were additionally chosen based on the availability of concurrent sediment chemistry and sediment bioassay data. The only exception was Station 12062 in Case Inlet (Fig. 1), which was included as a reference area for taxonomic analysis for use in the determination of the IBD Index, but for which sediment bioassay data were only available from later studies.

In conducting this study, it was recognized that the data sets were not completely compatible either temporally or spatially. Similarly, it was recognized that benthic nomenclature is not static, and that taxonomic identifications are liable to future revision. The intent of this study was to derive specific information from available data sets, without undertaking extensive additional sampling and analysis.

2.1 Geographical Study Areas

2.1.1 NMFS samples

NOAA samples represented collections by the National Marine Fisheries Service (NMFS). Five major embayments were considered in Puget Sound ranging from highly industrialized (Elliott Bay and the Duwamish, Commencement Bay Waterways, Sinclair Inlet) to non-industrialized areas (Case Inlet, Samish Bay) (Fig. 1; Table 1). Precise

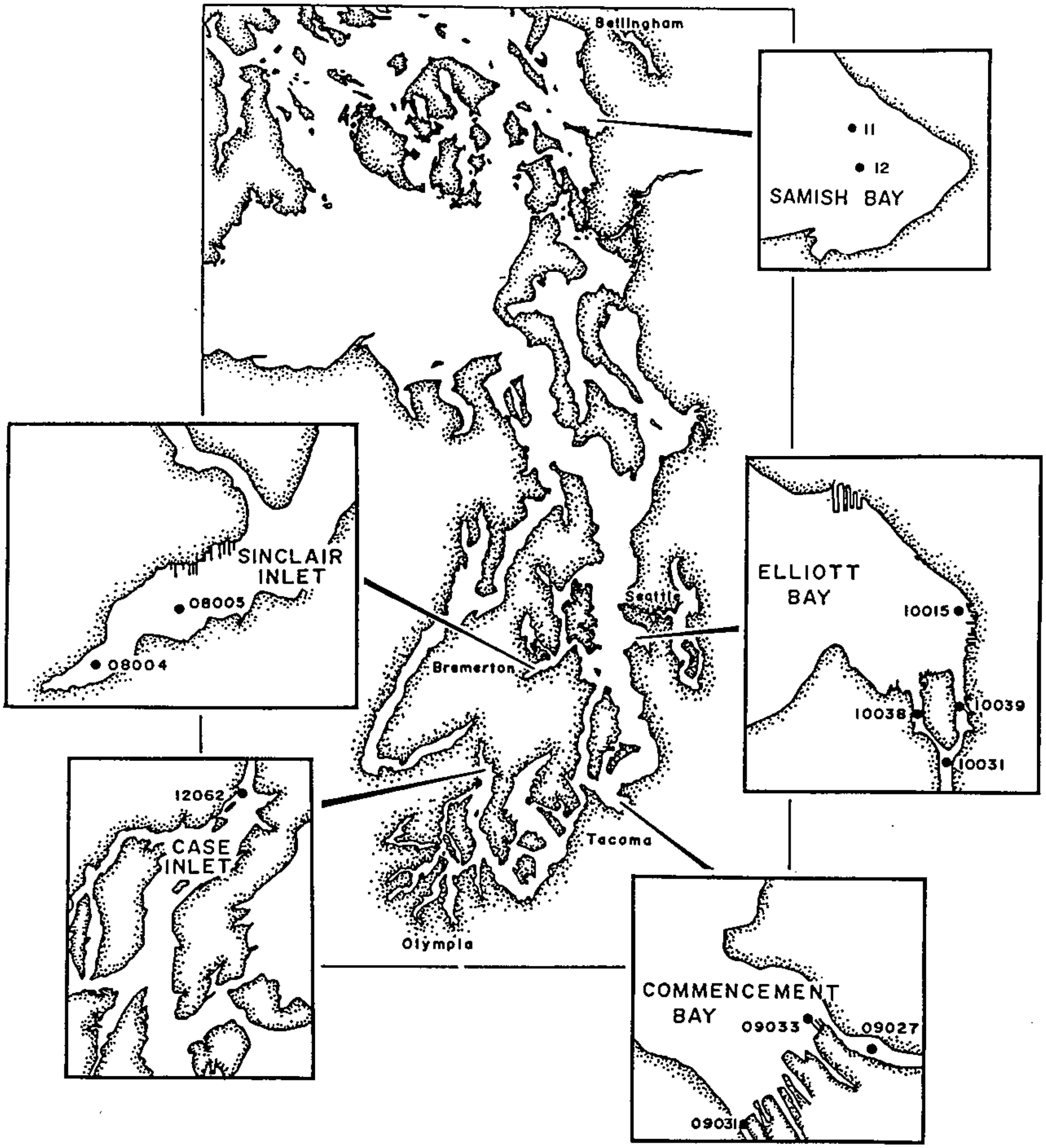

Figure 1. Location of Sampling Stations - NMFS Samples. Station numbers in Samish Bay are those of Chapman et al. (1984); all others are those of Malins et al. (1980,1982).

Table 1. Data relevant to NMFS stations used in this study

Location	Station numbers		Mean grain size (Phi)	Sand:mud ratio	Depth (m)	Bioassay responses[a]		
						Respiration	Genotoxicity	Oyster larvae
	Chapman et al., 1982a (sediment toxicity)	Malins et al., 1980 (sediment chemistry and infauna)						
ELLIOTT BAY								
Piers 54/56[b]	12	10015[e]	7.0	0.12	20-60	2	1	1
DUWAMISH								
W. Waterway[b]	20	10038	7.1	0.22	15-20	1	1	N.D.
E. Waterway[b]	25	10039	8.5	0.04	12-16	1	2	N.D.
S. Harbor Is.[b]	29	10031	7.1	0.18	10-20	2	2	2
COMMENCEMENT BAY								
Inner Bay[b]	40	09033	6.0	0.16	20-50	1	2	N.D.
Hylebos Waterway[b]	48	09027	6.9	0.03	10-12	2	2	N.D.
City Waterway[b]	67	09031	6.7	0.08	7-11	2	2	1
SINCLAIR INLET								
Inner[c]	76	08004	-	0.02	8-10	1	2	N.D.
Outer[b]	81	08005	-	0.03	10-12	2	1	N.D.
CASE INLET[b]	-	12062	7.4	0.06	20-45	N.D.	N.D.	N.D.
	Chapman et al., 1984							
SAMISH BAY								
A[d]	11	-	2.8	3.69	14	1	2	1
B[d]	12	-	6.0	0.10	15	1	1	1

a. N.D. = no data
Respiration 1 = no significant response; 2 = significant ($p<0.05$) response
Genotoxicity 1 = no significant reponse; 2 = significant ($p<0.05$) response
Oyster larvae 1 = no response; 2 = toxic response

b. Six samples quarterly in 1979; sampling by NMFS; taxonomy identifications by E.V.S. Consultants.

c. Six samples taken in three quarters in 1979 (not in February); sampling by NMFS, taxonomic identifications by E.V.S. Consultants.

d. Four samples taken once in 1983; sampling and taxonomic identifications by E.V.S. Consultants.

e. Benthos (less than 1 mm) not present in February 1979.

station location information is provided by Malins et al. (1980, 1982) for all areas except Samish Bay. Station location information for Samish Bay is provided by Chapman et al. (1984); these samples were collected in 1983 by E.V.S. Consultants.

2.1.2 Metro Seattle samples

Four stations located in the area of discharge for the Denny Way Combined Sewer Overflow (CSO) in Elliott Bay were included in this study (Fig. 2; Table 2). Precise station location information is available from the Municipality of Metropolitan Seattle (Metro).

2.1.3 EPA samples

Seven stations located in the Waterways of Commencement Bay were included in this study (Fig. 2; Table 3). Precise station locations are available from the U.S. EPA (Newport, Oregon, Marine Science Center).

2.2 Collection Methods and Taxonomic Identifications

2.2.1 NMFS samples

The following description of collection methodology is from Malins et al. (1980, 1982). Benthic samples were taken using a modified Van Veen grab (0.1 m^2). Two core samples each with a surface area of 100 cm^2 and a depth of 10 cm were taken from each grab for analysis of infaunal organisms. Three grab samples were taken at each station. Sediment from the 1000 cm^3 core samples was wet-sieved through 1 mm mesh stainless steel sieves and the material remaining was preserved, analyzed and then archived. Collections were undertaken quarterly during 1979 at a total of 37 stations in Puget Sound (and adjacent waters).

With the exception of samples from Samish Bay, all NMFS samples were collected in 1979 and processed as detailed above. Samples from Samish Bay were collected in May 1983 by E.V.S. Consultants using the same VanVeen grab. Two grab samples were taken at each station. Two 1000 cm^3 cores were taken from each grab using the NMFS core sampler, and treated as detailed above.

All taxonomic identifications reported for the NMFS samples were completed by E.V.S. Consultants as follows. Organisms were separated from archived samples under a Wild M3 or M5A stereomicroscope and placed into separate containers filled with alcohol according to the following major taxonomic groups: Amphipoda, Other Crustacea, Polychaeta, Oligochaeta, Mollusca, Nematoda and Others. All organisms were then identified to the lowest possible taxonomic level consistent with presently available literature. Identifications were done using either a Wild M5A stereomicroscope or a Leitz Laborlux compound microscope. All benthic samples, including reference species, are presently being stored by E.V.S. Consultants.

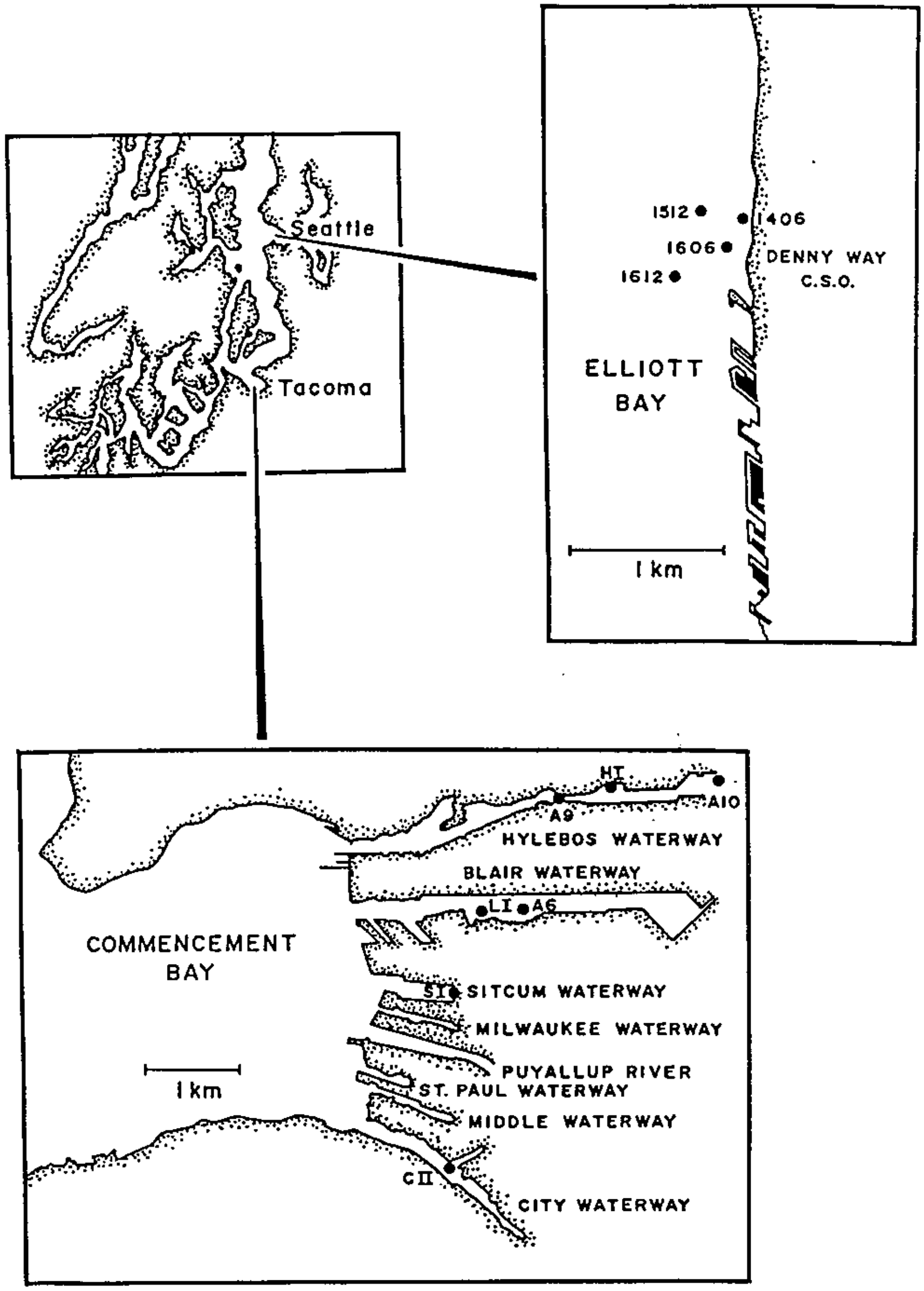

Figure 2. Location of Sampling Stations - EPA (Commencement Bay) and Metro Seattle (Elliot Bay) Samples. Station numbers are study specific.

Table 2. Data relevant to METRO stations used in this study

Station number[a]	Mean grain size (Phi)	Sand:mud ratio	Depth (m)	Amphipod bioassay result[b]	No. of Van Veen grabs used for benthic taxonomy
1406	5.8	0.23	19.3	1	1
1512	6.6	0.11	40.0	2	1
1606	5.6	0.23	30.1	2	1
1612	5.8	0.20	39.9	1	1

a. All stations from the area of the Denny Way CSO in 1982; sampling by METRO; taxonomic identifications by J. Word (Comiskey et al., 1983).

b. 1 = no response; 2 = toxic response

Table 3. Data relevant to EPA stations used in this study

Station number[a]	Mean grain size (Phi)	Sand:mud ratio	Depth (m)	Amphipod bioassay result[b]	No. of Van Veen grabs used for benthic taxonomy
Hylebos Waterway					
A9	7.0	0.15	13.0	1	1
A10	6.4	0.22	13.0	1	1
H1	7.3	0.06	13.0	2	5
Blair Waterway					
A6	6.4	0.18	18.0	2	1
L1	6.3	0.12	16.0	1	5
Sitcum Waterway					
S1	6.1	0.12	15.0	1	5
City Waterway					
C11	6.0	0.23	9.0	1	5

a. All stations from Commencement Bay Waterways in 1981; sampling and taxonomic identifications by EPA (Swartz et al., 1982).

b. 1 = no response; 2 = toxic response

2.2.2 Metro Seattle samples

The following description of methodology is from Comiskey et al. (1983). Benthic grab samples were collected in summer 1982 using a modified Van Veen grab ($0.1 m^2$). A minimum penetration depth of 10 cm was required in substrates ranging from silty and fine sand to silt. Samples (no subcoring) were screened through 1 mm mesh sieves and the material remaining was preserved and identified. A complete description of taxonomic methodologies is provided in Comiskey et al. (1983).

2.2.3 EPA samples

The following description of methodology was provided by Dr. R. Swartz (EPA Newport, pers. comm.). Benthic grab samples were taken in spring 1981 using a modified Van Veen grab ($0.1 m^2$). Samples (no subcoring) were screened through 1 mm mesh sieves and the material remaining was preserved and identified by EPA in-house staff. A complete description of taxonomic methodologies will be provided by Dr. R. Swartz in a forthcoming publication (Swartz, pers. comm.).

2.3 Computer Entry of Benthic Data

A total of 67 separate benthic samples (including seasonal samples at the same stations) were considered for inclusion in this study, representing a total of 23 stations in Puget Sound. Species names and the levels of taxonomic identification differed in some respects among the three data sets. Accordingly, best professional judgement was used to standardize the different data sets. The data were then tabularized on hard copy, keypunched and verified on computer systems at the University of Washington. All benthic data were standardized (by surface area) to numbers per $0.1 m^2$ of sediment.

The data were grouped into six samples for the control areas (four seasonal samples at Case Inlet and one sampling at each of two stations in Samish Bay). A total of 34 samples represented seasonal sampling conducted by NOAA at 8 stations in Elliott Bay, Commencement Bay Waterways and Sinclair Inlet. Four samples represented one-time sampling from four stations off the Denny Way CSO, conducted by Seattle Metro. Twenty-three samples represented one-time sampling from seven stations in Commencement Bay Waterways conducted by the U.S. EPA.

2.4 Application of the Index of Benthic Degradation (IBD)

O'Connor's (unpublished report) Index of Benthic Degradation (IBD) was applied to selected Puget Sound embayments using combined data from the NMFS, Metro Seattle and EPA data sets. Specific index definitions were amended as necessary following discussions with Dr. O'Connor regarding problems in interpretation or application. Population differences were determined based on percent contributions of the taxa. The following equation defines the Index.

$$I_{rc} = \text{antilog}\ (f_{rc})\cdot F_{rc}\cdot \sum_{i=1}^{n_{rc}} (n_{ir}\log(6 + \frac{0.05}{P_{irc}}))$$

The Index is intended for use in areas with different environmental strata, and in the above equation r = the r'th stratum, defined by natural environmental criteria. As only one environmental stratum was considered in the present study (depth and sediment characteristics being similar), r = 1. Two separate groups of stations (=substrata) were considered: reference areas (2 stations in Samish Bay, 1 in Case Inlet) and all other (pollution-affected) areas (20 stations in Elliott Bay, Commencement Bay Waterways and Sinclair Inlet).

The simplified Index used in the present study is as shown below:

$$I = \text{antilog}\ (f_c)x(F_c)x\Sigma[(n_i)\log(6 + {}^{0.05}/P_{irc})]$$

where f_c is the fraction of the total representative taxa that show significant population differences between control and study areas, F_c is the fraction of Phyla that contain taxa that show significant population differences between areas, n_i are the number of taxa that show significant population differences between areas at a selected confidence level and P_{irc} is the probability that differences are not significant (one minus the confidence level). Representative taxa are those present in sufficient abundance and frequency to allow reasonable analysis of spatial differences in their distributions.

In general usage typical values of the Index are expected to range between 0 and 1 if the degree of degradation between the control and study areas is slight and to range higher than 1 for increasing degradation. For example, the term within the summation [log (6 + $0.05/P_{irc}$)] is 0.813 for a 80% confidence level and increases to 1.014 for a 99% confidence level. The summation term will thus typically range from a low value of N_t x 0.813 to a high value of N_t x 1.014 where N_t is the total number of significant taxa at the lowest (i.e. species) level. The first term preceeding the summation sign, antilog (f_c), ranges in value between 1 and 10, whereas the value of the other term, F_c, must lie between 0 and 1. Thus, the value of the Index for less than 20% affected representative taxa, will be approximately 1.5 x F_c x N_t. Where N_t is three species or less, the value of the Index will be less than 1. For higher levels of affected taxa (in the range of 60 and 70%), the Index values increase to about 5 x F_c x N_t. If F_c is also in the 60% to 70% range and if the numbers of taxa that show significant population differences are fairly large (in the range of 30 or more) then the Index can exceed values of 100.

Instances of Index values greater than 100 were not considered by O'Connor (unpublished report) as they do not accord with the intent of the Index. The purpose of the Index is to provide a means to evaluate marginal degradation (O'Connor, pers. comm.). In such situations the number of taxa showing significant differences in occurrence would be few. Statistical concerns for such cases include a sufficient number of samples for both the control and the study areas, evaluation of the power of the test, and a correction for multiplicity of tests (in cases where due to large numbers of representative taxa, some differences may incorrectly be calculated as statistically significant). The latter statistical concern can be addressed by adjusting the level of confidence chosen for statistical significance.

In applying the Index to cases where marginal degradation is the concern, corrections for non-gaussian population distributions of the taxonomic variables are desirable, and a precise significance test such as the Student's t-test to determine the differences in population means would be the preferred approach. O'Connor (pers. comm.) has cautioned that in this application, the power of the significance of the tests must be calculated for all of the variables. If more than 30% of the taxa show a power that is insufficient (the probability of a Type 2 error of greater than 0.3), then the test that is being applied is not sufficient for the purpose.

In this particular study we were not concerned with marginal degradation, as the study areas are considered to be considerably degraded (c.f. Malins et al., 1980, 1982; Swartz et al., 1982; Comiskey et al., 1983). Because the study areas were more than marginally degraded, and because there were few reference samples for comparison, a more robust non-parametric significance test was chosen, in this case the Mann-Whitney U Test (Appendix D). In addition, the confidence level for determining significant differences in taxa was set at 99% (P=0.01).

The Mann-Whitney U Test is basically a calculation procedure which groups and compares all of the samples from one area versus all of the samples from a second area. If the population count for a taxon from the reference area is less than the population count for a taxon from the study area then a value of 1 is added to the U statistic. If the population count for a taxon from the reference area is greater than the population count for the same taxon from the study area, then a value of 0 is added to the U statistic. This calculation procedure is repeated for all combinations of taxa from both the control and the study areas. If there is no difference in the mean value of the population for that taxonomic variable between the reference area and the study area, then the U statistic will have a value close to the mean between the two possible extremes. However, if the population counts are significantly shifted between the two groups, then the U statistic will tend toward very low values (if the population counts for the reference area are greater on the average than the population counts for the study area), or high values (if the population counts for the study area are greater than the population counts for the reference area). Critical values for the U statistic are then chosen based on the confidence level desired for determining a significant population difference between the two areas.

The Mann-Whitney U Test was applied to three groups of representative taxa selected in a empirical iterative process on the basis of frequency of abundance and average occurrence criteria. The least severe criteria selected taxa with abundances greater than zero in 10% of the samples or an average occurrence of greater than 2.0 individuals/m^2. These two criteria were initially chosen to ensure that ecologically sensitive (and hence potentially rare) taxa (i.e., phoxocephalid amphipods) would not be overlooked or discriminated against by criteria levels that were too high. The selection of initial criteria was in accord with the fact that the limited reference data set consisted of only six samples. If a taxon were only observed in each of these six samples, over the entire data set (51 samples) that would represent a frequency of occurrence of 11.8%. Application of these criteria to the benthic population data available for this study resulted in the selection of 79 representative taxa. Application of more stringent criteria, namely that the frequency of occurrence be greater than zero in at least 20% of the samples or that the average occurrence when observed be greater than or equal to 5.0 individuals/m^2 resulted in the selection of 47 representative taxa. A third set of selection criteria, that the frequency of occurrence be greater than zero in at least 20% of the samples and that the average abundance when observed be greater than or equal to 1.0 individuals/m^2 resulted in the selection of 19 representative taxa.

The Mann-Whitney U Test was applied to each of the three groups of representative taxa and used to calculate the resultant Index values for each group. In addition to the Index values based on all of the benthic data, calculations of the Index were undertaken separately for taxa from Commencement Bay Waterways and Elliott Bay (plus the Duwamish), using the 79 representative taxa.

2.5 Correlations Between Sediment Chemistry, Benthic Communities and Sediment Bioassays

Broad-scale sediment bioassay, chemistry and community structure data for the three data sets were different in many respects. Sediment bioassay data for the NMFS samples which had chemical and benthic infauna data included three tests: oligochaete respiration anomalies, fish cell genotoxicity and oyster larvae toxicity (Table 1). Sediment bioassay data for the Metro and EPA samples included only amphipod acute lethality (Tables 2, 3).

Sediment chemistry data included parameters that were common to all samples and parameters measured in only one or two of the three data sets (Table 4). Differences in nomenclature were resolved internally for benthic infauna community structure data prior to analysis; however, as per the chemistry data, some species were common to all data sets while others were recorded in only one or two of the three different studies.

Multivariate data analysis techniques contained in the pattern recognition system "Arthur" were used to evaluate relationships among the benthic population data and between the benthic data and the available chemical data. The Arthur software system is described in Quinlan et al. (in press). The system contains multivariate data

Table 4. Description of available sediment chemistry data

	NMFS samples[a]	Metro Seattle samples[b]	EPA samples[c]
Common To All Samples	General grain size TOC TVS Pollution-related metals Organics PAHs PCBs	General grain size TOC TVS Pollution-related metals Organics PAHs PCBs	General grain size TOC TVS Pollution-related metals Organics PAHs PCBs
Not Common To All Samples	Non-anthropogenic metals Organics Pesticides Chlorobutadiene	Non-anthropogenic metals Organics Pesticides Hexachlorobutadienes Phthalate esters Other organic compounds	Iron and manganese General oil/grease sulfides ammonia Organics Chlorinated pesticides Chlorobutadiene Phthalate esters

a. Data from Malins et al., 1980.
b. Data from Comiskey et al., 1983.
c. Data from Schults et al., unpublished manuscript.

analysis routines for cluster analysis, for factor analysis, for determining the significance of the importance of variables in distinguishing between groups of samples, and for classification and modeling methodologies. For this particular study the techniques of most interest were as follows: techniques to identify variables that showed significant or potentially significant differences between groups of samples; techniques for cluster analysis; techniques for factor analysis (used to show relationships between the variables); the technique for the Pearson Linear correlation calculation (for evaluating linear relationships between variables); and, techniques for multivariate regression analysis (to help evaluate the relationships between variables).

The analysis techniques within the Arthur system were applied in various ways. Initial exploratory data analysis was done on the benthic population data alone using factor analysis, cluster analysis and correlation techniques. The data were evaluated comparing the results among the four geographical locations (Commencement Bay Waterways, Elliott Bay and the Duwamish, Sinclair Inlet and reference stations), calculating the variance weight differences for each of the variables between these groups. Selected feature plots were completed based on the results observed in the variance weight calculations. The benthic data were then merged with available chemical data for the stations, and exploratory techniques using factor analysis and factor analysis plotting were applied along with the Pearson Linear correlation to calculate the correlations between each of the variables.

Sediment bioassay data patterns were compared with the results of the above statistical analyses. This comparison involved subjective rather than statistical criteria due to major differences in the data for the studies used in the analysis. Benthic community structure was categorized based on the total proportion of polychaetes and molluscs present (species characteristic of degraded environments), and the presence or absence of phoxocepholid amphipods (species sensitive to degraded environments). Sediment toxicity was categorized based on sediment bioassay results. Sediment contaminant levels were characterized based on pooled concentrations of three representative metals (Pb, Cu, Zn), and on pooled concentrations of combustion PAHs and of PCBs.

3.0 RESULTS

3.1 Taxonomic Identifications

3.1.1 NMFS samples

A listing of all benthic invertebrates identified from the archived NMFS samples examined is provided in Table 5 and a summary of the abundance data is provided in Table 6 including the mean number of each taxon for each station. Complete data tables for each sample by station and season are provided in Appendix A.

Table 5. Benthic invertebrates identified from NMFS samples

Taxon
Coelenterata
Hydrozoa
Anthomedusae
Corynidae
Sarsia
Leptomedusae
Anthozoa
Pennatulacea
Pennatulidae
Ptilosarcus gurneyi
Nemertea
Nematoda
Sipunculida
Sipunculoidea
Golfingiidae
Golfingia
Annelida
Oligochaeta
Tubificida
Tubificidae
Limnodriloides victoriensis
Annelida
Polychaeta
(Errantiate)
Polynoidae
Glyceridae
Glycera americana
G. capitata
G. robusta
Sigalionidae
Pholoe minuta
Sthenelais tertiaglabra
Phyllodocidae
Anaitides sp.
A. groenlandica
A. hartmanni
Eteone (Mysta) sp.
E. barbata
E. californica
E. spilotus
Eulalia sp.
E. levicornuta
Mystides sp.
Paranaitis polynoides
Pilargidae
Pilargis berkleyae
Sigambra tentaculata
Syllidae
Eusyllinae
Exogone molesta
Sphaerosyllis brandhorsti
Typosyllis harti
Nereidae
Nereis procera
Nephtyidae
Nephtys cornuta cornuta
N. cornuta franciscana
N. ferruginea
Sphaerodidae
Sphaerodoropsis sphaelifer
Goniadidae
Goniada brunnea
G. maculata
Glycinde armigera
G. picta
Onuphidae
Diopatra ornata
Nothria sp.
N. elegans
N. iridescens
Lumbrineridae
Lumbrineris sp.
L. bicirrata
L. californiensis
L. cruzensis
L. luti
Dorvilleidae
Schistomeringos sp.
S. annulata
S. caeca

Table 5 (continued)

Hesionidae
Ophiodromus pugettensis
Podarkeopsis (=*Gyptis*) *brevipalpa*

Annelida
Polychaeta
(Sedentariate)
Orbiniidae
Leitoscoloplos pugettensis
Scoloplos acmeceps

Paraonidae
Aricidea minuta
A. ramosa
A. suecica
Levinsenia (=*Tauberia*) *gracilis*

Magelonidae
Magelona berkleyi
M. longicornis

Chaetopteridae
Spiochaetopterus costarum

Spionidae
Laonice cirrata
Polydora caulleri
P. socialis
Prionospio cirrifera
P. (=*Paraprionospio*) *pinnata*
P. steenstrupi
Spiophanes sp.
S. berkleyorum

Cirratulidae
Chaetozone setosa
Cirratulus cirratus
Tharyx sp.
Tharyx sp. B
T. multifilis

Cossuridae
Cossura soyeri

Opheliidae
Armandia brevis
Ophelina breviata

Sternaspidae
Sternaspis scutata

Capitellidae
Barantolla americana
Capitella sp.
C. capitata
Heteromastus filobranchus
Mediomastus sp.
M. ambiseta
M. californiensis
Notomastus lineatus
N. tenuis

Maldanidae
Microclymene acirrata
Praxillella affinis
P. gracilis
Rhodine bitorquata

Oweniidae
Owenia fusiformis

Amphictenidae
Pectinaria californiensis

Scalibregmidae
Scalibregma inflatum

Terebellidae
Artacama conifera
Pista brevibranchiatus
P. cristata
Polycirrus sp.
P. californicus
Proclea graffii

Trichobranchidae
Lanassa venusta venusta
Terebellides stroemi

Amphitritinae

Ampharetidae
Amage anops
Ampharete sp.
A. arctica (=*finmarchica*)
Amphicteis sp.
A. scaphobranchiata
Anobothrus sp.
A. gracilis

Sabellidae
Euchone limnicola
E. incolor

Phoronida

Mollusca
Gastropoda
Pyramidellida
Pyramidellidae

Table 5 (continued)

Odostomia quadrae
Turbonilla escholtzei

Cephalaspidea
Cylichnidae
Cylichna alba

Gastropteridae
Gastropteron pacifica

Neogastropoda
Columbellidae
Amphissa columbiana
Mitrella permodesta
M. tuberosa

Nassariidae
Nassarius fraterculus

Archaeogastropoda
Turbinidae
Moelleria quadrae

Mesogastropoda
Naticidae
Polinices pallidus

Rissoidae
Alvania compacta
Rissoina newcombi

Bivalvia
Myoida
Myidae
Sphenia ovoidea

Nuculoidea
Nuculidae
Acila castrensis
Nucula tenuis

Nuculanidae
Nuculana minuta

Pholadomyoida
Cuspidariidae
Cardiomya pectinata

Lyonsiidae
Lyonsia arenosa

Pandoridae
Pandora filosa

Mytiloidea
Mytilidae
Rhomboidella (=*Megacrenella*) *columbiana*

Veneroida
Cardiidae
Nemocardium centifilosum

Cooperellidae
Cooperella subdiaphana

Kelliidae
Kellia suborbicularis

Lucinidae
Lucina (=*Parvilucina*) *tenuisculpta*
Lucinoma annulata

Montacutidae
Mysella
M. tumida

Solenidae
Sole sicarius

Tellinidae
Macoma calcarea
M. carlottensis
M. elimata
M. nasuta
M. yoldiformis
Tellina carpenteri
T. modesta

Veneridae
Compsomyax subdiaphana
Protothaca staminea

Thyasiridae
Axinopsida serricata

Echinodermata
Ophiuroidea
Ophiurida
Amphiuridae
Amphiodia
A. urtica

Ophiuridae
Ophiura luetkeni

Holothuroidea
Dendrochirotida
Phyllophoridae
?*Havelockia* ?*benti*

Table 5 (continued)

Cucumaridae
Cucumaria piperata

Arthropoda
Crustacea
Decapoda
Anomura
Porcellanidae

Brachyura
Pinnotheridae
Pinnixa
P. eburna
P. schmitti

Xanthidae
Lophopanopeus bellus bellus

Thalassinida
Callianassidae
Callianassa
C. californiensis

Caridea
Crangonidae
Crangon alaskensis

Amphipoda
Caprellidea
Gammaridea
Phoxocephalidae
Harpiniopsis
Heterophoxus
Paraphoxus

Lysianassidae
Lysianassa (=*Aruga*)
Orchomene pinguis

Corophiidae
?*Microdeutopus*

Cumacea
Leuconidae
Eudorella pacifica

Diastylidae
Diastylis

Myodocopida
Philomedidae
Ephilomedes carcharodonta
E. producta

Cylindroleberididae

Cirripedia
Balanidae
Balanus crenatus

Mysidacea
Mysidae
Xenacanthomysis pseudomacropsis
Neomysis

Tanaidacea
Paratanaidae
Leptochelia

Table 6. Summary of benthic taxa present in NMFS samples. Total numbers found and (in parentheses) the number of samples in which they were found

Taxon	Elliott Bay + Duwamish 10015[a]	Elliott Bay + Duwamish 10031[b]	Elliott Bay + Duwamish 10038[b]	Elliott Bay + Duwamish 10039[b]	Sinclair Inlet 08004[a]	Sinclair Inlet 08005[b]	Commencement Bay 09027[b]	Commencement Bay 09031[b]	Commencement Bay 09033[b]	Case Inlet 12062[b]	Samish Bay 11[c]	Samish Bay 12[c]
Coelenterata										2(1)		
Hydrozoa												
Anthomedusae												
Corynidae												
Sarsia					2(2)							
Leptomedusae										2(1)		
Anthozoa												
Pennatulacea												
Pennatulidae												
Ptilosarcus gurneyi	1(1)	1(1)						1(1)				1
Nemertea	7(2)	4(2)	2(1)	1(1)	1(1)				2(2)	16(3)	1	
Nematoda									6(1)			
Sipunculida												
Sipunculoidea												
Golfingiidae												
Golfingia											1	1
Annelida												
Oligochaeta												
Tubificida												
Tubificidae												
Limnodriloides victoriensis	1(1)										1	
Annelida												
Polychaeta												
(Errantiate)												
Polynoidae									1(1)			1
Glyceridae								1(1)				
Glycera americana		1(1)	4(2)			3(1)		1(1)	3(1)	1(1)		1
G. capitata	7(2)		2(1)			1(1)	1(1)	5(2)	6(1)		2	
G. robusta												1
Sigalionidae												
Pholoe minuta		7(3)	3(2)	1(1)	2(1)	8(3)	4(3)	1(1)		18(4)	3	4
Sthenelais tertiaglabra											1	

Table 6 (continued)

Taxon	Elliott Bay + Duwamish 10015a	10031b	10038b	10039b	Sinclair Inlet 08004a	08005b	Commencement Bay 09027b	09031b	09033b	Case Inlet 12062b	Samish Bay 11c	12c
Phyllodocidae												
Anaitides sp.							2(1)					
A. groenlandica									6(3)			
A. hartmanni		10(4)					1(1)		2(2)			
Eteone (Mysta) sp.		2(1)			1(1)		1(1)					
E. barbata										1(1)		
E. californica		6(3)	1(1)	4(3)	2(1)	1(1)	6(4)	1(1)	6(3)			
E. spilotus	1(1)				1(1)				1(1)			
Eulalia sp.									1(1)			
E. levicornuta	1(1)											
Mystides sp.			1(1)						1(1)			
Pacanaitis polynoides											1	
Pilargidae												
Pilargis berkleyae									1(1)			1
Sigambra tentaculata										7(4)		
Syllidae												
Eusyllinae	1(1)								1(1)			
Exogone molesta									1(1)			
Sphaerosyllis brandhorsti							2(1)		1(1)			
Typosyllis harti	2(1)											
Nereidae												
Nereis procera					1(1)				1(1)	1(1)		
Nephtyidae												
Nephtys cornuta cornuta									2(1)	1(1)		
N. cornuta franciscana					1(1)	2(2)	2(1)	9(4)	6(1)	5(2)	33	4
N. ferruginea	4(2)		3(1)			2(1)	1(1)	7(3)	18(3)	1(1)	1	
Sphaerodidae												
Sphaerodoropsis sphaelifer	2(1)							2(2)	6(1)			
Goniadidae												
Goniada brunnea	1(1)								1(1)			
G. maculata							1(1)					
Glycinde armigera			1(1)					1(1)		1(1)		1
G. picta		1(1)			3(1)		1(1)			3(2)		
Onuphidae												
Diopatra ornata								1(1)	1(1)			
Nothria sp.									1(1)			
N. elegans												1
N. iridescens	3(2)								1(1)			

Table 6 (continued)

Taxon	Elliott Bay + Duwamish 10015a	10031b	10038b	10039b	Sinclair Inlet 08004a	08005b	Commencement Bay 09027b	09031b	09033b	Case Inlet 12062b	Samish Bay 11c	12c
Lumbrineridae												
Lumbrineris sp.		1(1)	1(1)		1(1)	2(1)			1(1)			
L. bicirrata			1(1)									1
L. californiensis		3(1)	2(1)							2(1)		
L. cruzensis		1(1)	6(3)	19(1)	4(2)	39(4)			1(1)	12(3)	4	3
L. luti	20(3)	116(4)	41(4)	49(3)	6(3)	25(4)	45(4)	15(2)	79(4)		2	6
Dorvilleidae												
Schistomeringos sp.		1(1)										
S. annulata		2(2)							1(1)			
S. caeca						1(1)	4(3)					
Hesionidae												
Ophiodromus pugettensis								2(1)		5(2)		1
Podarkeopsis (=*Gyptis*) *brevipalpa*					1(1)	3(1)	1(1)	1(1)	1(1)	5(2)		
Annelida												
Polychaeta												
(Sedentariate)												
Orbiniidae												
Leitoscoloplos pugettensis										2(1)	1	
Scoloplos acmeceps									1(1)	3(1)		
Paraonidae												
Aricidea minuta										3(2)		
A. ramosa										1(1)		
A. suecica	5(1)			1(1)			1(1)	1(1)	3(2)	1(1)		
Levinsenia gracilis	6(3)			1(1)		1(1)			1(1)		7	9
Magelonidae												
Magelona berkleyi										1(1)		
M. longicornis										1(1)		3
Chaetopteridae												
Spiochaetopterus costarum			2(1)			5(1)				2(2)		
Spionidae												
Laonice cirrata	4(2)			1(1)					2(1)		2	2
Polydora caulleri								1(1)	5(2)			
P. socialis		1(1)		1(1)	2(1)	3(2)			4(2)			
Prionospio cirrifera	1(1)	1(1)	3(2)		2(1)		3(1)	43(4)	1(1)	26(3)	1	1
P. (=*Paraprionospio*) *pinnata*			2(1)		50(3)	17(4)	1(1)	2(1)	2(2)	17(3)		
P. steenstrupi	7(3)	2(1)	2(1)			13(4)	2(1)	7(2)	11(3)	10(1)		
Spiophanes sp.						1(1)			2(1)	1(1)		
S. berkleyorum					8(3)	3(2)			1(1)	7(2)		

Table 6 (continued)

Taxon	Elliott Bay + Duwamish 10015a	10031b	10038b	10039b	Sinclair Inlet 08004a	08005b	Commencement Bay 09027b	09031b	09033b	Case Inlet 12062b	Samish Bay 11c	12c
Cirratulidae												
Chaetozone setosa	3(2)	5(3)	20(4)	5(4)	28(3)	10(4)	9(4)	4(2)	11(3)			1
Cirratulus cirratus		395(4)	4(2)				3(1)					
Tharyx sp.								1(1)				
Tharyx sp. B			2(1)		3(1)	2(1)	1(1)	2(1)	4(3)			1
T. multifilis		581(4)	19(1)	167(4)	305(3)	53(4)	81(4)	7(4)	77(4)	1(1)		
Cossuridae												
Cossura soyeri			3(2)		7(2)	4(2)	14(3)	61(3)	1(1)	2(1)		
Opheliidae												
Armandia brevis		5(1)			1(1)		1(1)	4(3)			2	
Ophelina breviata												1
Sternaspidae												
Sternaspis scutata											1	2
Capitellidae												
Barantolla americana								1(1)	5(2)			
Capitella sp.									2(1)			
C. capitata		17(3)	5(2)				2(2)	3(3)	3(1)			
Heteromastus filobranchus	68(3)	31(4)	36(4)	12(4)	2(2)	1(1)	10(4)	47(4)	91(4)		1	
Mediomastus sp.		2(2)				1(1)	1(1)	5(1)		23(2)		
M. ambiseta								1(1)		19(1)		
M. californiensis	3(2)	3(2)	1(1)					2(1)	2(2)	1(1)		
Notomastus lineatus									1(1)			
N. tenuis	6		7(4)					4(1)	70(4)			1
Maldanidae	5(3)								2(2)			
Microclymene acirrata	29(3)			1(1)		7(1)						
Praxillella affinis				1(1)			1(1)	1(1)	60(4)		2	3
P. gracilis	7(3)						1(1)	11(4)	95(4)			1
Rhodine bitorquata	1(1)											
Oweniidae												
Owenia fusiformis											1	3
Amphictenidae												
Pectinaria californiensis									1(1)	1(1)		
Scalibregmidae												
Scalibregma inflatum					15(2)	11(2)			1(1)			
Terebellidae		1(1)		1(1)					11(1)			
Artacama conifera									8(3)			
Pista brevibranchiatus											1	
P. cristata						1(1)			12(2)			
Polycirrus sp.	1(1)		1(1)		3(3)	1(1)			5(3)	1(1)		
P. californicus	3(1)							3(1)				
Proclea graffii	2(1)											

Table 6 (continued)

Taxon	Elliott Bay + Duwamish 10015[a]	10031[b]	10038[b]	10039[b]	Sinclair Inlet 08004[a]	08005[b]	Commencement Bay 09027[b]	09031[b]	09033[b]	Case Inlet 12062[b]	Samish Bay 11[c]	12[c]
Mesogastropoda												
Naticidae												
<u>Polinices pallidus</u>										1(1)		
Rissoidae												
<u>Alvania compacta</u>							23(3)					
<u>Rissoina newcombi</u>								1(1)				
Bivalvia		1(1)					1(1)	1(1)				
Myoida												
Myidae												
<u>Sphenia ovoidea</u>			1(1)									
Nuculoidea												
Nuculidae												
<u>Acila castrensis</u>						45(4)	1(1)	1(1)	1(1)			
<u>Nucula tenuis</u>	8(2)		8(3)	1(1)		6(2)		2(1)	46(4)		1	1
Nuculanidae												
<u>Nuculana minuta</u>	2(2)		1(1)							1(1)		
Pholadomyoida												
Cuspidariidae												
<u>Cardiomya pectinata</u>	3(2)		1(1)									
Lyonsiidae												
<u>Lyonsia arenosa</u>	4(2)	2(2)	1(1)	1(1)	2(2)				14(2)	2(1)		
Pandoridae												
<u>Pandora filosa</u>		1(1)										
Mytiloidea												
Mytilidae												
<u>Rhomboidella</u> (=<u>Megacrenella</u>) <u>columbiana</u>			2(2)						3(2)			
Veneroida												
Cardiidae												
<u>Nemocardium centifilosum</u>	3(1)								5(3)			
Cooperellidae												
<u>Cooperella subdiaphana</u>			5(1)			4(1)		1(1)				
Kelliidae												
<u>Kellia suborbicularis</u>		3(1)		1(1)	37(3)	49(3)	1(1)	7(3)		4(2)		1
Lucinidae												
<u>Lucina</u> (=<u>Parvilucina</u>) <u>tenuisculpta</u>											1	
<u>Lucinoma annulata</u>	2(1)						1(1)	2(2)	1(1)	1(1)		

Table 6 (continued)

Taxon	Elliott Bay + Duwamish 10015[a]	10031[b]	10038[b]	10039[b]	Sinclair Inlet 08004[a]	08005[b]	Commencement Bay 09027[b]	09031[b]	09033[b]	Case Inlet 12062[b]	Samish Bay 11[c]	12[c]
Trichobranchidae												
Lanassa venusta venusta		29(3)					1(1)	2(2)	10(2)	1(1)		
Terebellides stroemi	2(1)				14(3)	14(3)		1(1)				
Amphitritinae					1(1)							
Ampharetidae												
Amage anops									1(1)			
Ampharete sp.									1(1)			
A. arctica (=_finmarchica_)	1(1)											
Amphicteis sp.					1(1)							
A. scaphobranchiata					1(1)	2(1)						
Anobothrus sp.										1(1)		
A. gracilis									1(1)			
Sabellidae												
Euchone limnicola		100(3)	6(1)			1(1)		1(1)				
E. incolor					1(1)			5(2)				
Phoronida												
Mollusca												
Gastropoda												
Pyramidellida												
Pyramidellidae												
Odostomia quadrae						1(1)	1(1)					
Turbonilla escholtzei												1
Cephalaspidea												
Cylichnidae												
Cylichna alba										1(1)		
Gastropteridae												
Gastropteron pacifica	1(1)						1(1)					
Neogastropoda												
Columbellidae												
Amphissa columbiana							1(1)					
Mitrella permodesta			1(1)		2(1)	3(2)	2(1)					
M. tuberosa									1(1)			
Nassariidae												
Nassarius fraterculus										1(1)		
Archaeogastropoda												
Turbinidae												
Moelleria quadrae											1	

Table 6 (continued)

Taxon	Elliott Bay + Duwamish 10015a	10031b	10038b	10039b	Sinclair Inlet 08004a	08005b	Commencement Bay 09027b	09031b	09033b	Case Inlet 12062b	Samish Bay 11c	12c
Montacutidae												
Mysella					3(1)	1(1)						
M. tumida					47(3)	4(2)	3(2)	13(4)	2(2)	1(1)		
Solenidae												
Sole sicarius		1(1)										1
Tellinidae												
Macoma calcarea					1(1)	1(1)			1(1)		2	
M. carlottensis	22(2)	30(4)	46(4)	72(4)	6(3)	9(4)	2(1)	127(4)	666(4)	4(2)		
M. elimata		1(1)	4(2)						20(4)			
M. nasuta							1(1)	10(1)		1(1)		
M. yoldiformis								15(1)		1(1)		
Tellina carpenteri					2(1)							
T. modesta					1(1)			3(1)				
Veneridae												
Compsomyax subdiaphana						3(2)			6(2)	1(1)		
Protothaca staminea								1(1)				
Thyasiridae									1(1)			
Axinopsida serricata	120(3)	56(4)	129(4)	150(4)	25(3)	123(3)	126(4)	200(4)	1013(4)	7(3)		
Echinodermata										1(1)		
Ophiuroidea												
Ophiurida												
Amphiuridae						2(1)		3(2)	8(3)	3(3)		
Amphiodia									17(1)	147(4)	23	26
A. urtica									3(1)	52(4)	8	3
Ophiuridae									8(1)			
Ophiura luetkeni									1(1)			
Holothuroidea												
Dendrochirotida												
Phyllophoridae												
?*Havelockia* ?*benti*												1
Cucumaridae												
Cucumaria piperata						1(1)						
Arthropoda												
Crustacea												
Decapoda						1(1)			1(1)			
Anomura								2(2)				
Porcellanidae						1(1)						

Table 6 (continued)

Taxon	Elliott Bay + Duwamish				Sinclair Inlet		Commencement Bay			Case Inlet	Samish Bay	
	10015a	10031b	10038b	10039b	08004a	08005b	09027b	09031b	09033b	12062b	11c	12c
Brachyura	1(1)				4(1)			4(1)	1(1)			
Pinnotheridae			1(1)		8(1)						69	86
Pinnixa					6(2)	10(2)		9(2)	2(2)	3(1)		
P. eburna			1(1)	1(1)		5(1)		10(2)				
P. schmitti					1(1)	3(2)		10(2)		6(3)	11	6
Xanthidae												
Lophopanopeus bellus bellus										1(1)		
Thalassinida												
Callianassidae									1(1)			
Callianassa								6(2)				
C. californiensis								6(2)				
Caridea												
Crangonidae												
Crangon alaskensis					1(1)							
Amphipoda												
Caprellidea					2(1)							
Gammaridea										1(1)	7	2
Phoxocephalidae												
Harpiniopsis											7	6
Heterophoxus										2(2)	2	2
Paraphoxus										1(1)	2	2
Lysianassidae												
Lysianassa (=*Aruga*)												1
Orchomene pinguis											1	
Corophiidae												
?*Microdeutopus*												2
Cumacea					25(1)	1(1)		1(1)	14(1)			
Leuconidae						17(1)		5(1)				
Eudorella pacifica		1(1)			3(1)	45(3)	1(1)	22(2)	2(1)	4(2)	31	14
Diastylidae												
Diastylis									1(1)			
Myodocopida	3(1)	4(2)		2(1)	1(1)		2(2)	23(2)				
Philomedidae												
Ephilomedes carcharodonta	2(1)	61(3)	16(2)	3(2)			13(2)	121(2)	95(4)		2	31
E. producta	2(1)			1(1)			6(1)	22(2)	124(4)		9	
Cylindroleberididae												2
Cirripedia												
Balanidae												
Balanus crenatus					9(1)							

Table 6 (continued)

Taxon	Elliott Bay + Duwamish				Sinclair Inlet		Commencement Bay			Case Inlet	Samish Bay	
	10015[a]	10031[b]	10038[b]	10039[b]	08004[a]	08005[b]	09027[b]	09031[b]	09033[b]	12062[b]	11[c]	12[c]
Mysidacea												
Mysidae												
Xenacanthomysis pseudomacropsis							1(1)					
Neomysis							1(1)					
Tanaidacea												
Paratanaidae												
Leptochelia			1(1)									

a. Number of organisms collected in 6 samples (2 subsamples of each of 3 Van Veen grabs) collected on each of 3 occasions in 1979. Numbers in parentheses indicate number of sampling occasions (1, 2 or 3) on which the taxa were present.

b. Number of organisms collected in 6 samples (2 subsamples of each of 3 Van Veen grabs) collected on each of 4 occasions in 1979. Numbers in parentheses indicate number of sampling occasions (1, 2, 3 or 4) on which the taxa were present.

c. Number of organisms collected in 4 samples (2 subsamples of each of 2 Van Veen grabs) collected once in 1982.

3.1.2 Metro Seattle samples

Complete data tables are presented in Appendix B.

3.1.3 EPA samples

Complete data tables are presented in Appendix C.

3.1.4 Combined NMFS, Metro and EPA data

Table 7 presents data on the relative occurrence of selected taxa in NMFS benthic samples. These data indicated that at stations considered to be impacted by pollution (Elliott Bay, Duwamish Waterway, Sinclair Inlet, Commencement Bay Waterways), the benthic community was composed predominantly of polychaetes and molluscs (73.2-98.4% of totals). Arthropods and echinoderms were generally not an important part of the community at these stations. In addition, the pollution-sensitive amphipod families Phoxocephalidae and Lysianassidae were generally absent from these stations. In contrast, at the reference stations (Case Inlet and Samish Bay), the importance of polychaetes and molluscs in the benthic community dropped dramatically (23.8-47.0% of totals). Echinoderms and arthropods were more abundant, and the sensitive amphipod families Phoxocephalidae and Lysianassidae were present at the reference stations.

A similar pattern was evident for the Metro Seattle and EPA samples (Table 8), which were all from stations considered polluted. Polychaetes and molluscs comprised the majority of the fauna (84.4-99.2% of totals). Echinoderms and arthropods were generally not an abundant faunal component. Phoxocephalidae were present only in very low abundances (0.7% of total) in Elliott Bay, while Lysianassidae were present only in very low abundances at three of the seven Commencement Bay Waterways stations (maximum 0.2% of total faunal composition).

3.2 Application of the Index of Benthic Degradation (IBD)

As described in Section 2.4, three separate selection criteria were used in an empirical, iterative process to derive taxa for calculation of the Index. The first, least restrictive criteria included 79 taxa, the second included 47 taxa, and the third included 19 taxa. The Index was calculated separately for each taxa grouping. The program used for application of the Mann-Whitney U statistic is shown in Appendix D.

Six higher-order taxonomic groupings (for the "F" parameter) were identified in the initial 79 taxa grouping: Nemertea, Nematoda, Annelida, Mollusca, Echinodermata and Arthropoda. Four of these (Annelida, Mollusca, Echinodermata and Arthropoda) contained taxa significantly different (P=0.01) in populations between the reference and test stations, producing an "F" value of 0.667. The number of representative taxa with a significant population difference at the 99% confidence level was 64. This produced an "f" fraction of 0.797. The Index, calculated using these fraction terms together with the term

Table 7. Relative occurrence of selected taxa in NMFS samples

Area:	Elliott Bay	Duwamish River			Sinclair Inlet		Commencement Bay			Case Inlet	Samish Bay	
Location:	Pier 54/56	S. Harbor Is.	W. W'way	E. W'way	inner	outer	Hylebos	City	inner	Reach Is.		
Station:	10015	10031	10038	10039	08004	08005	09027	09031	09033	12062	11	12
Polychaeta	51.8[a]	88.9	44.9	53.1	71.2	41.5	52.0	30.2	24.2	41.3	27.2	21.8
Lumbrineridae	5.2	8.1	20.2	13.7	1.7	11.5	11.5	1.7	3.0	3.0	2.4	4.1
Spionidae	3.1	0.3	1.0	0.4	9.5	6.5	1.5	5.9	1.0	13.4	1.2	1.2
Cirratulidae	0.8	65.8	11.3	34.6	51.2	11.3	24.0	1.6	3.4	0.2	0	0.8
Capitellidae	20.2	3.6	12.2	2.4	0.3	0.3	3.3	7.0	6.4	9.5	0.4	0
Maldanidae	1.0	0	0	0.4	0	1.2	0.5	1.3	5.8	0	0.8	1.6
Others	21.5	11.1	0.2	1.6	8.5	10.7	11.2	12.7	4.6	15.2	22.4	14.1
Mollusca	42.9	6.4	49.9	45.3	19.2	43.5	41.8	43.0	65.3	5.7	2.0	2.0
Axinopsida serricata	31.4	3.8	32.3	30.2	3.8	21.5	32.1	22.6	37.2	1.5	0.4	0
Macoma carlottensis	5.8	2.0	11.5	14.5	0.9	1.6	0.5	14.2	24.4	0.9	0	0
Macoma nasuta	0	0	0	0	0	0	0.3	1.5	0	0.5	0	0
Others	5.7	0.6	6.1	0.6	14.5	20.4	8.9	4.7	3.7	2.8	1.6	2.0
Polychaeta & Mollusca	94.7	95.2	94.8	98.4	90.4	85.0	93.8	73.2	89.5	47.0	29.2	23.8
Echinodermata	0	0	0	0	0	0.5	0	0.3	1.4	44.6	12.6	12.3
Arthropoda	2.1	4.4	4.8	1.4	9.1	14.5	6.1	26.9	8.8	4.0	57.3	63.1
Phoxocephalidae	0	0	0	0	0	0	0	0	0	0.9	7.3	4.9
Lysianassidae	0	0	0	0	0	0	0	0	0	0	0.4	0.4
Others	2.1	4.4	4.8	1.4	9.1	14.5	6.1	26.9	8.8	3.1	49.6	57.8

a. Percent occurrence; values greater than 50% are underlined.

Table 8. Relative occurrence of selected taxa in METRO and EPA samples

	METRO	EPA						
Area:	Elliott Bay	Commencement Bay						
Location:	Denny Way CSO[a]		Hylebos			Blair	Sitcum	City
Station:	1406 + 1512 + 1606 + 1612	A9	H1	A10	L1	A6	S1	C11
Polychaeta	29.5[b]	88.8	92.9	63.9	35.7	49.9	52.5	70.9
Lumbrineridae	2.7	7.5	5.7	5.8	3.2	5.1	0	<0.1
Spionidae	1.8	0.8	0.4	0.8	1.0	1.6	0.7	14.9
Cirratulidae	1.5	77.9	79.9	50.4	23.5	17.7	40.4	15.0
Capitellidae	4.3	0.2	0.2	0.4	1.8	1.5	0.7	1.4
Maldanidae	2.5	0.4	< 0.1	0.4	1.6	8.7	< 0.1	0
Others	16.7	2.0	6.6	6.1	4.6	15.3	10.6	39.5
Mollusca	54.8	8.1	6.3	35.4	58.2	43.6	45.8	21.9
Axinopsida serricata	33.3	1.2	3.0	34.7	51.7	36.0	23.5	0.4
Macoma carlottensis	13.8	0	0	0	0	0	0	0
Macoma nasuta	0	0.6	1.0	0.8	2.1	3.3	16.5	5.8
Others	7.7	6.3	2.3	0.1	4.4	4.3	5.8	15.7
Polychaeta & Mollusca	84.4	96.9	99.2	99.3	93.9	93.5	98.3	92.8
Echinodermata	< 0.1	0	< 0.1	0	0.2	1.2	0.2	0
Arthropoda	15.2	3.1	0.8	0.7	5.0	5.5	1.3	7.0
Phoxocephalidae	0.7	0	0	0	0	0	0	0
Lysianassidae	0	0.2	< 0.1	0	< 0.1	0	0	0
Others	14.5	2.9	0.7	0.7	4.9	5.5	1.3	7.0

a. Mean of the four stations.
b. Mean percent occurrence; values greater than 50% are underlined.

based on the probabilities and the number of taxa significant at that probability, produced a resulting IBD value of 304.8. Details of the calculation are shown below.

$$IBD = I_{rc} = \text{antilog}(f_c)\cdot F_c \cdot \sum_{i=1}^{n_{12}} [(n_i)\cdot \log(6 + {}^{0.05}/_{P_{irc}})]$$

$$= (6.273)(0.667)[7(\log(6.25)) + 2(\log(7)) + 63(\log(11))]$$

$$= (6.273)(0.667)[5.571 + 1.690 + 65.608]$$

$$= (6.273)(0.667(72.844) = \underline{\underline{304.8}}$$

In the case of the 47 representative taxa derived by the second, more restrictive criteria, the "F" fraction remained at 0.667. Of the 47 representative taxa, 35 showed a significant (P=0.01) population difference between the control and the study areas resulting in an "f" fraction of 0.745. The corresponding IBD value was calculated to be 152.8 as shown below.

$$IBD = I_{rc} = \text{antilog}(fc)\cdot F_c \cdot \sum_{i=1}^{n_{12}} [(n_i)\cdot \log(6 + {}^{0.05}/_{P_{irc}})]$$

$$= (5.55)(0.667)[5(\log(6.25)) + 1(\log(6.5)) + 35(\log(11))]$$

$$= (5.55)(0.667)[3.98 + 0.813 + 36.45]$$

$$= (3.702)[41.24] = \underline{\underline{152.8}}$$

For the 19 representative taxa derived by the third and most restrictive criteria, the IBD calculation was based on four higher-order groupings (Annelida, Mollusca, Echinodermata and Arthropoda). All four of these contained taxa significantly different (P=0.01) in populations between the reference and test stations, producing an "F" fraction of 1.0. Fifteen of the 19 representative taxa had significant (P=0.01) population differences, resulting in an "f" fraction of 0.789. The corresponding IBD value was calculated to be 96.0 as shown below.

$$IBD = I_{rc} = \text{antilog}(fc)\cdot F_c \cdot \sum_{i=1}^{n_{12}} [(n_i)\cdot \log(6 + {}^{0.05}/_{P_{irc}})]$$

$$= \text{antilog}(0.789)\cdot 1.0\cdot [(15)\log(11)]$$

$$= (6.15)(1.0)(15)(1.041)$$

$$= \underline{\underline{96.0}}$$

Following calculation of the Index by the above three criteria levels for all samples, the Index was recalculated for Commencement Bay Waterways and Elliott Bay separately, using the least restrictive criteria which included 79 taxa. Overall differences in the relative abundances of the benthic taxa between the reference areas and the two urban areas are shown in Table 9, and served to derive the "F" and "f" values. The Index value for Commencement Bay Waterways was calculated to be 162, while that for Elliott Bay was calculated to be 107. The principal difference between the two areas lay in the

Table 9. Numbers of benthic taxa found to be different between Commencement Bay Waterways and Elliott Bay using the IBD calculation

Taxonomic grouping	Commencement Bay Waterways						Elliott Bay (and the Duwamish)					
P[a] =	n.s.	0.2	0.1	0.05	0.02	0.01	n.s.	0.2	0.1	0.05	0.02	0.01
Nermertea	1[b]	-	-	-	-	-	-	1	-	-	-	-
Nematoda	1	-	-	-	-	-	1	-	-	-	-	-
Annelida	9	2	1	2	4	30	15	1	3	1	5	23
Mollusca	1	-	-	1	5	9	2	-	1	-	4	9
Echinodermata	-	-	-	-	-	3	1	-	-	-	-	2
Arthropoda	3	1	1	-	1	4	5	-	-	-	2	3
TOTALS	15	3	2	3	10	46	24	2	4	1	11	37

a. P = level of significance; n.s. = not significant.

b. Number of taxa; 79 total taxa for each of Commencement Bay Waterways and Elliott Bay.

fact that more Annelida were significantly different from the reference area in Commencement Bay Waterways than in Elliott Bay. The corresponding "F" values were 0.667 for both urban areas (4 of 6 higher order groupings significant). The remaining differences between the two areas were slight. The corresponding "f" values for Commencement Bay Waterways (46 significantly different representative taxa) and Elliott Bay (37 significantly different representative taxa) were 0.582 and 0.468, respectively (Table 9).

A detailed examination of the differences in taxa abundances between Commencement Bay Waterways and Elliott Bay revealed a total of 14 taxa with significantly different relative distributions in each embayment compared to the reference area (Table 10). Commencement Bay Waterways had ten taxa with significantly greater abundances than observed in the reference areas, in contrast to Elliott Bay which had none. Two species, Pholoe minuta and Pinnixa schmitti, were present in significantly lower abundances in Elliott Bay compared to the reference areas. One genus, Heterophoxus, was present in significantly lower abundances in Commencement Bay Waterways compared to the reference areas. The substantially greater IBD value for Commencement Bay Waterways compared to Elliott Bay is thus primarily due to greater abundances of particular taxa in Commencement Bay Waterways, rather than to an absence of taxa.

3.3 Correlations Between Sediment Chemistry, Benthic Communities and Sediment Bioassays

The multivariate analyses involved initial application to the benthic community data and subsequent combined application to the benthic and sediment chemistry data. Inclusion of sediment bioassays was a third comparative step that did not involve multivariate statistics. The results of the first two steps are reviewed herein, while the overall, non-statistical comparison is discussed in Section 4.3. Hard copy of all computer runs discussed here are available for review through E.V.S. Consultants.

3.3.1 Benthic communities

A variety and combination of techniques within the Arthur system were used to explore and illustrate relationships in the benthic population data. An initial exploratory factor analysis run on benthic population data suggested specific groupings of the benthic taxa which showed similar distribution trends. The taxa contributing most heavily to the first five factors (=groupings) are identified in Table 11. Two factors in particular contained groupings that could be differentiated as pollution sensitive or non-sensitive taxa. Factor two was loaded positively with taxa such as the phoxocephalid amphipod genus Heterophoxus which are considered to be sensitive to increasing pollution levels, and it is loaded negatively with two taxa, one of which (Lumbrineris luti) is considered to be more tolerant to pollution. In contrast, factor five is loaded negatively with taxa such as the Amphiodia that are considered to be sensitive to pollution.

Table 10. Significantly different taxa between Elliott Bay and Commencement Bay Waterways

Taxa	Commencement Bay Waterways		Elliott Bay (and the Duwamish)	
	U-statistic value[a]	P =[b]	U-statistic value[a]	P =[b]
Pholoe minuta	77	n.s.	6	0.01
Prionospio cirrifera	97	0.02	30	0.1
Tharyx sp.	108	0.01	57	n.s.
Cossura soyeri	101	0.01	51	n.s.
Praxillella affinis	99	0.01	67	n.s.
Scalibregma inflatum	106	0.01	57	n.s.
Artacama conifera	108	0.01	57	n.s.
Euchone incolor	109	0.01	57	n.s.
Macoma nasuta	104	0.01	48	n.s.
Amphiuridae	100	0.01	38	n.s.
Decapoda	109	0.01	57	n.s.
Pinnixia schmitti	60	n.s.	12	0.01
Callianassa californiensis	108	0.01	57	n.s.
Heterophoxus	28	0.1	96	0.02

a. U Statistic $U_{exp} = 57$ $U_{ma} = 114$ $U_{min} = 0$
U > 57, study average > control average
U < 57, control average < study average.

b. P = level of significance; n.s. = not significant.

Table 11. Initial exploratory factors based on all representative taxa

Factor	Positive loadings	Negative loadings
1	Axinopsida serricata Nucula tenuis, Nephtys ferruginea Macoma carlottensis Amphiuridae Macoma eliminata Heteromastus filobranchus, Artacama conifera, Praxillella gracilis Lyonsia arenosa	Praxillella affinis, Anaitides groenlandica
2	Levinsenia gracilis Heterophoxus Nephtys cornuta franciscana Eudorella pacifica Pinnixia, Pinnixia schmitti	Lumbrineris luti, Laonice cirrata
3	Odostomia quadrae, Turbonilla escholtzei Tharyx sp., Euchone incolor Decapoda Scalibregma inflatum	Prionospio pinnata Kellia suborbicularis
4	Scalibregma inflatum, Prionospio pinnata Mysella tumida Pinnixia Podarkeopsis brevipalpa	Anaitides hartmanni Cirratulus cirratus
5	Terebellides stroemi Rhomboidella columbiana Proclea graffii	Mediomastus sp. Amphiodia urtica, Amphipodia Prionospio cirrifera

Thus, both factor two in a positive direction and factor five in a negative direction appear to reflect sensitivity to pollution levels based on the species which are loading heavily onto those factors.

A factor plot (Fig. 3) supports this interpretation, and provides a visual qualitative separation of data sets and stations. The lower right-hand corner of Figure 3, corresponding to high abundances of factor two organisms and high abundances of factor five organisms, includes stations with the highest population densities of the most pollution sensitive species, and therefore should reflect the least amount of degradation due to pollution. This corner does include the samples from the reference areas. The Hylebos Waterway station (HI) to the furthest left of Figure 3, with the least value on factor two, exhibits exactly opposite behavior to that evidenced by the two Samish Bay stations. Additional information derivable from this plot includes separation of the Denny Way CSO samples into a reasonably distinct group. Samish Bay samples are distinct from Case Inlet samples, and appear to represent less degraded conditions. In general, the NMFS samples and the EPA samples are distributed in the same area of the plot, indicating both a similar degree of pollution effect on the benthos, and that the data sets are comparable. Finally, although there are some differences within the NMFS samples related to different sampling times at the same stations (i.e. the four Case Inlet stations), these differences are generally not major.

A more complete exploratory run was done on the 79 representative taxa derived as described in Section 2.4. Variance weights were calculated to compare distributional differences between the reference and study areas. Variance weights in excess of 2.0 indicate a significant difference. The following three taxa showed significant differences between the two groups of stations, as indicated by the variance weights: Pholoe minuta, 2.83; Amphiodia, 2.97; Amphiodia urtica, 2.17. Mean variance weights for these three taxa were also considerably (5x) greater in the reference areas compared to the study areas.

Factor analysis was applied to the benthic data in this run, and the results of the first five factors (including retained variance due to each factor) are provided in Table 12. Pholoe minuta is not included within these factors and appears to act differently from the other taxa. Factors two and three are plotted against each other in Figure 4. In general, this plot is similar to Figure 3. The bulk of the samples show a similar diffuse behavior, with two exceptions. The Samish Bay samples and Hylebos station HI are clearly separated from each other and from all other stations. The positioning of these stations on the plot (Fig. 4) indicates that Samish Bay and station HI behave oppositely in relation to factor two. For factor three, Samish Bay behaves similarly to the bulk of the stations, whereas station HI behaves differently. The Case Inlet samples behaved similarly to the rest of the study area samples for these two factors.

Cluster analysis of the data showed the same general pattern. In general, the dendograms yielded similar results to Figure 3.

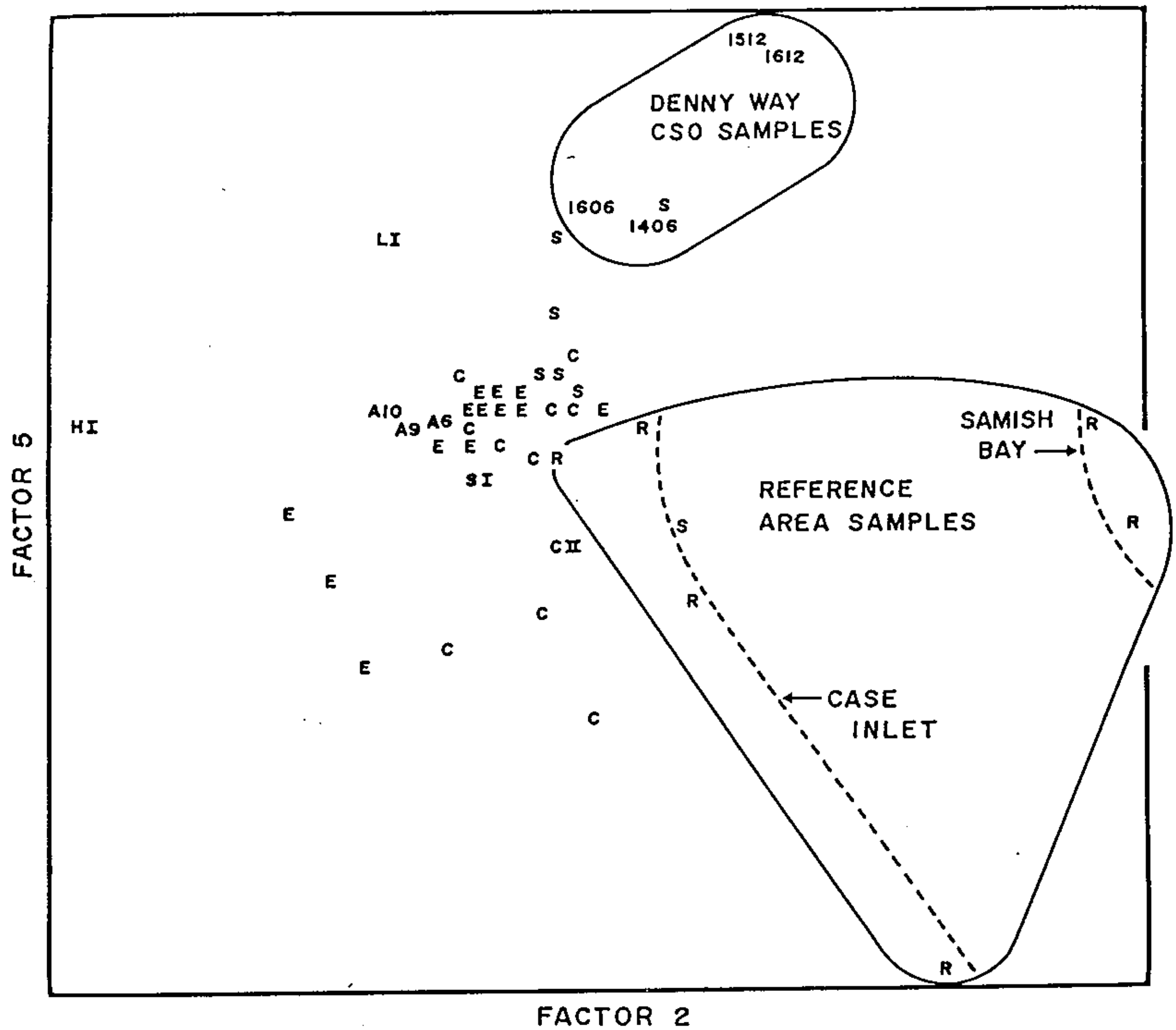

Figure 3. Factor plot of all benthic data. Factor loadings are identified in Table II. Boundaries are drawn by eye to provide qualitative information. Samples taken by Seattle Metro off Denny Way and by the U.S. EPA in Commencement Bay are identified by station number. NMFS samples are identified by location: R = reference, S = Sinclair Inlet, C = Commencement Bay Waterways, E = Elliott Bay and the Duwamish.

Table 12. Factor loadings for 79 representative taxa

Factor	(variance retained)	Positive loadings	Negative loadings
1	(15.23%)	Axinopsida serricata Nucula tenuis Nephtys ferruginea Praxillella affinis Anataides groenlandica Macoma carlottensis Amphiuridae Macoma elimata Heteromastus filobranchus Artacama conifera Praxillella gracilis Lyonsia arenosa	
2	(12.84%)	Levinsenia gracilis Nematoda Nephtys cornuta franciscana Lucinidae Limnodriloides verrucosus Laonice cirrata Eudorella pacifica Euphilomides producta Heterophoxus Pinnixa schmitti Pinnotheridae	
3	(8.83%)	Tharyx sp. Turbonilla escholtzei Decapoda Euchone incolor Odostomia quadrae Lumbrineris luti	Prionspio pinnata Kellia suborbicularis Scalibregma inflatum
4	(7.50%)	Anataides hartmanni Cirratulus cirratus Capitella capitata Euchone limnicola Lanassa venusta venusta	Prionospio steenstrupi Podarkeopsis brevipalpa Pinnixa Cossura soyeri Prionospio pinnata
5	(7.05%)	Mediomastus sp. Amphiodia urtica Prionospio cirrifera Amphiodia Podarkeposis brevipalpa	Proclea graffi Rhomboidella columbiana Terebellides stroemi Compsomyax subdiaphana Lumbrineris sp.

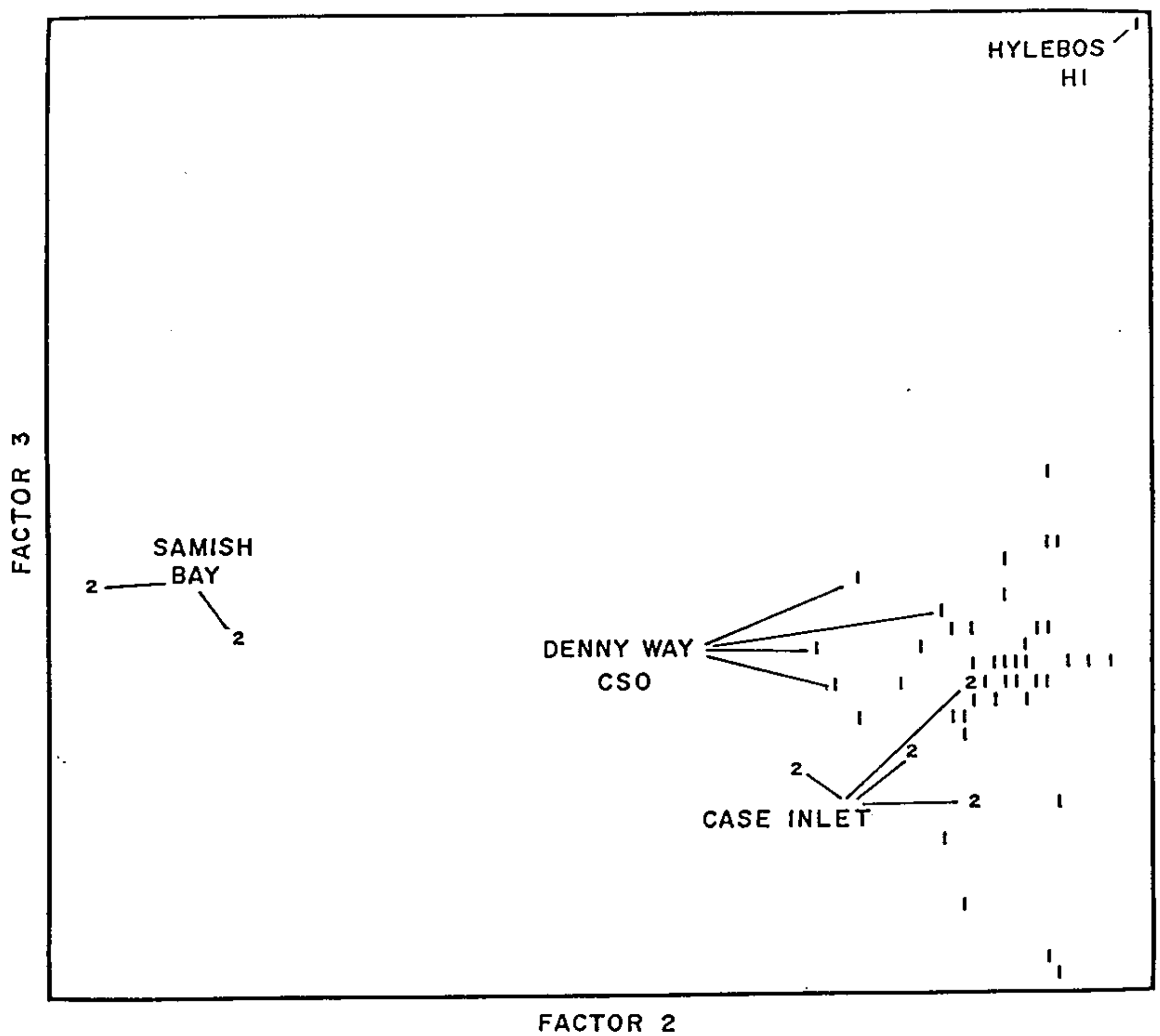

Figure 4. Factor plot of 79 representative taxa. Factor loadings are identified in Table 12. 1 = study area samples, 2 = reference area samples.

Following these analyses, the samples were then broken down into four categories for more detailed evaluation of differences: Elliott Bay (and the Duwamish), Sinclair Inlet, Commencement Bay Waterways, and the reference samples (Case Inlet and Samish Bay). Variance weights were calculated to compare these station groupings. On the basis of the number of taxa with significant variance weights and the size of those variance weights, there appear to be significant differences in species distributions between Elliott Bay and the reference area and Elliott Bay and Sinclair Inlet, but not as much difference between Elliott Bay and Commencement Bay Waterways. Thus, not only are there differences between the reference area and the study area as a whole, but there are differences between subsets of the study area, and those subsets also have significant differences from the reference area.

Based on indications from the variance weights, selected feature-feature plots were run comparing distributions of particular taxa. The results of these plots supported the results of previous analyses, indicating separation of the reference area samples (particularly Samish Bay) from the study area samples, and suggesting differences in species distributions within the study area samples.

3.3.2 Benthic communities and sediment chemistry

Initial attempts to evaluate relationships between the benthic infaunal and chemical data encountered serious problems. First, as detailed in similar analyses conducted by Quinlan et al. (in press), chemical data from different studies showed distinct groupings and correlations that indicated possible methodological biases, i.e. samples were grouped by laboratory. Second, differences in compounds analyzed and the detection limits used varied between the different studies. For instance, the samples analyzed by NMFS were characterized by extensive organic analyses to low detection limits, but metals data were derived from methods not compatible with those of other laboratories (Quinlan et al., in press). For these reasons, it was not possible to combine chemical analytical data for all samples. Attempts to characterize benthic communities in terms of sediment chemistry therefore concentrated on the organics data for the NMFS data sets which included the reference sample stations, and encompassed the greatest spatial area. Although the compatibility of some of the chemical identifications has been questioned (c.f. Quinlan et al., in press), for the purposes of the present study these data were used as provided.

The Pearson-linear correlation evaluation technique contained within the Arthur system was used along with factor analysis to examine relationships between sediment chemistry and benthic communities. Significant (P=0.01) correlations were found for several of the chemical and taxonomic variables. Some of these relationships appeared to reflect dependence on grain size. For instance, larger grain size was positively correlated with distributions of *Prionospio pinnata*, *Chaetozone setoza*, *Scalibregma inflatum*, *Terebellides stroemi*, and *Kellia suborbicularis*. Small grain size was negatively correlated with Decapoda.

Total organic carbon (TOC) was positively correlated with most of the taxa that were also correlated with small grain size: *Prionospio pinnata*, *Scalibregma inflatum*, *Terebellides stroemi*, *Mysella tumida*, and *Kellia suborbicularis*. This pattern reflects the innate correspondence between TOC and grain size.

Many of the polycyclic aromatic hydrocarbons (PAHs) showed positive correlations with four taxa: *Heteromastus filobranchus*, Maldanidae, *Artacama suecica*, and *Proclea graffi*. Hexachlorobenzene showed a different pattern, not correlating to these four taxa, but rather to eight separate taxa. Particularly high correlations among these eight taxa were noted for *Tharyx* sp. (0.951), *Euchone incolor* (0.971), *Odostomia quadrae* (0.883), and *Turbonilla escholtzei* (0.856). Lindane was positively correlated with 18 taxa. Particularly high correlations were noted for *Nucula tenuis* (0.910) and *Axinopsida serricata* (0.858). Aldrin showed a similar pattern to Lindane. These results appear to reflect the fact that both the chemicals and the taxa involved were present at only a few stations; correlations based on a small number of samples are sensitive to rare occurrences. In addition, the above results should be considered in light of the fact that measurements of non-PCB chlorinated hydrocarbons in Puget Sound sediments to date are considered unreliable (Quinlan et al., in press).

The DDT group showed a few negative correlations and fewer positive correlations. PCBs showed a considerable number of positive correlations (*Eteone californica*, *Cirratulus cirratus*, *Lumbrineris luti*, *Tharyx multifilis*, *Capitella capitata*, *Lanassa venusta venusta*, *Euchone limnicola*) and a few negative correlations (*Amphiodia*, *Amphiodia urtica*, *Pinnixa schmitti*).

Factor analysis was run on the chemistry and benthos data to determine whether similar behavior could be determined for specific station groupings. The first factor (accounting for 26.9% of the variance retained) consisted of negative loadings for PAHs, a similar pattern to that determined by Quinlan et al. (in press). The second factor loading (17.3%) was a mixture of negative loadings for hexachlorobenzene, lindane, aldrin and 11 taxa. Factors 3 (13.9%) and 4 (10.3%) evidenced grain size effects, together with a mixture of loadings for both chemicals and taxa. In general, these results reinforced the linear correlation analyses, indicating possibilities of relationships between some of the organic pollutant variables and benthic populations.

A plot of factors 1 and 2 is presented in Figure 5. This plot shows a separation of the four major study areas: Elliott Bay, Sinclair Inlet, Commencement Bay Waterways, and the reference areas. Samish Bay and Case Inlet are distinguishable from each other. In the Elliott Bay grouping, station 10015 (Seattle waterfront) was separated from the three Duwamish River stations because of much higher PAH loadings. Commencement Bay Waterway stations were less obviously separable.

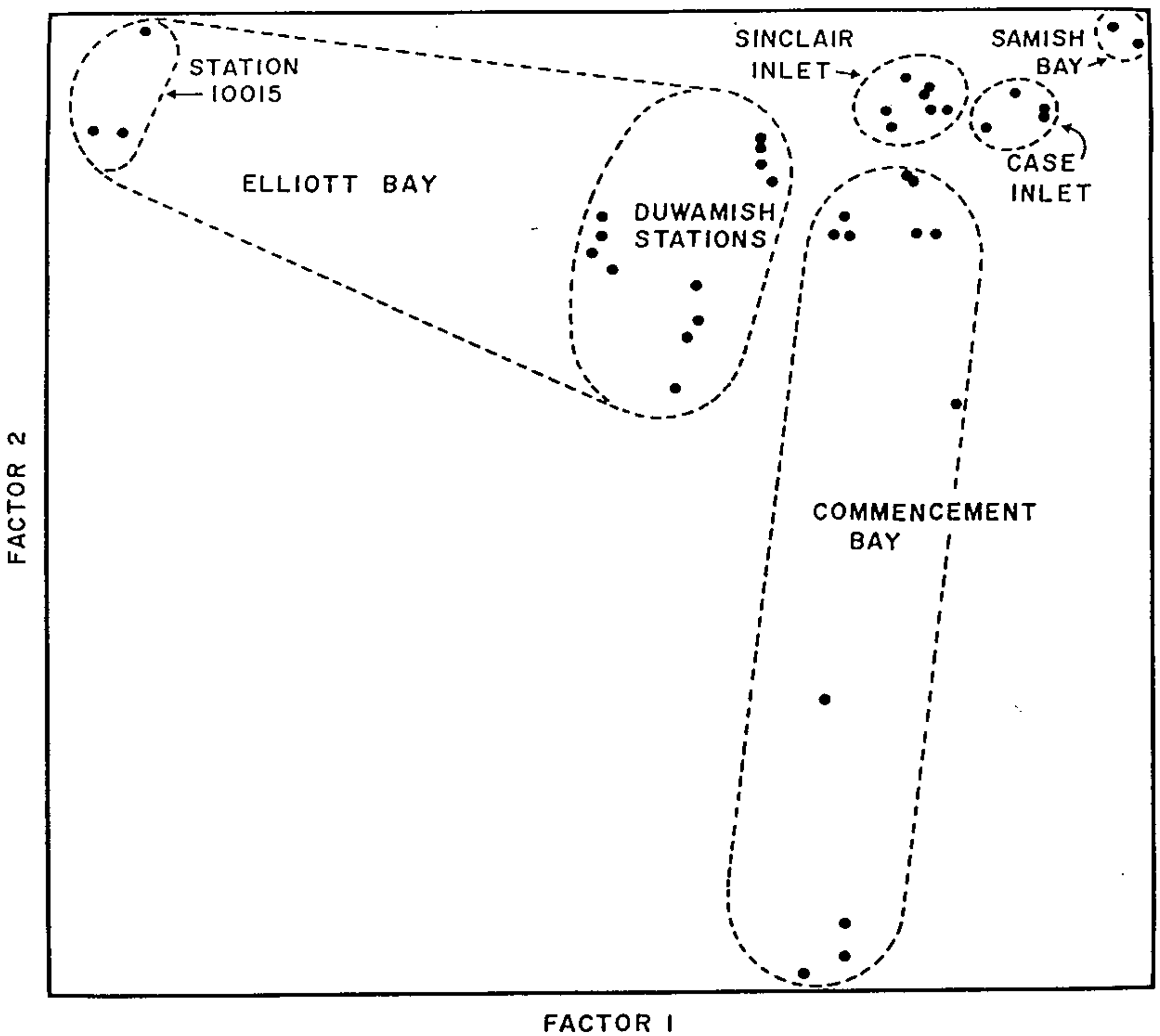

Figure 5. Benthos-chemistry factor plot by sample indices. Boundaries are drawn by eye. Factor 1 = PAHs. Factor 2 = hexachlorobenzene, lindane, aldrin, Axinopsida serricata, Nucula tenuis, Nephtys ferruginea, Anataides groenlandica, Praxillella affinis, Praxillella gracilis, Macoma carlottensis, Amphuiridae, Macoma elimata, Artacama conifera.

The factor plot (Fig. 5) indicates that the Commencement Bay Waterways can generally be distinguished based on high values of factor 1, whereas Elliott Bay tends towards high values of factor 2. The reference areas are low in values for both factors, with values for Samish Bay being lower than those for Case Inlet.

The results of this factor plot generally support the results of analyses conducted on the benthos alone (Section 3.3.1). For instance, Samish Bay was the cleanest, least pollution-affected reference area, and seasonal sampling at the same stations, e.g. Case Inlet, generally did not indicate major seasonal differences over a one year period. Inclusion of chemistry with benthic data permitted a separation of Elliott Bay, Sinclair Inlet and Commencement Bay Waterways, a pattern which was only suggested by analysis of the benthic community data alone.

4.0 DISCUSSION

4.1 Taxonomic Identifications

Identification of the NMFS samples and comparison with similar data sets from the U.S. EPA and the Municipality of Metropolitan Seattle showed a high level of agreement in terms of species present in particular areas, and dominant groups. A very high proportion of polychaetes and molluscs was found at all stations from urban embayments, in contrast to the three reference stations. This observation is in accord with data for other urban areas such as Bellingham Bay where the infauna is composed, on average, of 86% polychaetes and molluscs (Benedict et al., 1984). In the New York Bight, polychaetes and molluscs comprise an average of 95% of the benthic infauna (Caracciolo and Steimle, 1983).

The absence or low abundance of echinoderms and crustaceans from all but the reference stations is characteristic of areas with large waste inputs or residues (Pearson and Rosenberg, 1978). Caracciolo and Steimle (1983) noted that in the New York Bight amphipods are generally intolerant of pollutants and are virtually absent from polluted areas. The absence of sensitive phoxocephalid amphipods from all urban areas other than the Denny Way CSO is of significance. Swartz et al. (1982) found that phoxocephalid amphipods were absent from Commencement Bay Waterways, where high levels of sediment toxicity were recorded. Oakden et al. (1984) have shown that Rhepoxynius spp. exhibit a behavioral avoidance response to contaminated sediments. These authors noted that: ". . . sublethal effects on habitat selection may be as important in establishing and maintaining field populations as the mortality caused by a relatively polluted substrate."

Because of temporal incompatibilities between the data sets (collection dates: NMFS, 1979; EPA, 1981; Metro, 1982; Samish Bay, 1983), it was not possible to identify possible species-specific trends in abundance. Nichols (in press) has shown that long-term changes occur in Puget Sound benthic infauna as a result of year-to-year variations.

Similarly, Santos and Bloom (1983) have shown that natural changes in benthic infaunal species composition occur over periods ranging from months to decades.

4.2 Application of the Index of Benthic Degradation (IBD)

Application of the Index to the study areas, using 79 representative taxa, produced a value of 304.8. Reductions in the numbers of representative taxa first to 47 and then to 19 produced values of 152.8 and 96.0, respectively. Only this latter value was within the 0-100 range described by O'Connor (unpublished report). A value of 100 is presently described as the maximum for this index, which is intended for areas of marginal degradation, not for severely degraded areas (O'Connor, pers. comm.). On the basis of the IBD values calculated for combined data from Elliott Bay, Commencement Bay Waterways and Sinclair Inlet as compared to Case Inlet and Samish Bay, the former three embayments can be classified as substantially degraded in terms of benthic infaunal populations compared to the two reference embayments. This analysis corresponds to previous knowledge of the status of these areas (Malins et al., 1980, 1982; Swartz et al., 1982; Comiskey et al., 1983).

Comparative calculation of the Index for Elliott Bay compared to Commencement Bay Waterways indicated that these two areas were similar. This comparison points out the fact that the IBD may prove useful both for indicating areas of marginal degradation (the original intent), and for comparisons between more grossly degraded areas (the present application). This latter application allowed highlighting of specific differences between Elliott Bay (and the Duwamish) and Commencement Bay Waterways. If more samples were available, particularly from reference areas, a more rigorous statistical comparison would be possible, allowing finer discrimination of differences between areas based on individual taxa. Such an application would permit both assessment of the relative degree of change in one area versus another in terms of reference areas, and assessment of the particular differences that are important to that degree of change.

A previous application of the IBD was attempted by Diaz and Reid (1984). These authors succeeded in calculating Index values for marginally degraded areas in the New York Bight (range 7.5 to 21.7) using the Chi-Square test rather than the Mann-Whitney U test. As in the present study, the numbers of reference stations available were inadequate. These authors concluded that the Index was useful and workable but that there was a need to ". . . streamline its calculation." We concur with these conclusions with the following additions. First, "streamlining" should involve a better explanation of the purpose and limitations of the Index, particularly as demonstrated in this study. Second, the possibility of using the Index to compare degraded areas should be emphasized and explored.

4.3 Correlations Between Sediment Chemistry and Benthic Communities

Multivariate analyses were used to differentiate stations and areas based on benthic community data, and combined benthic community and sediment chemistry data. Analyses of the benthos alone allowed separation of the reference stations from the urban stations, and indicated that Samish Bay stations were less degraded than Case Inlet stations. It was not possible to statistically differentiate different urban embayments, although there were indications of such a separation based upon the benthos data.

Analyses of the combined benthos and chemistry data, restricted to the NMFS samples, confirmed the trends noted for benthos alone and allowed for separation of each embayment based on sediment PAH levels versus sediment levels of hexachlorobenzene, lindane, aldrin and the distribution of a few benthic taxa. Elliott Bay, Commencement Bay Waterways, Sinclair Inlet, Case Inlet and Samish Bays all emerged as distinct groupings in relation to these discriminating factors, particularly the PAH levels. In addition, two distinct groupings were determined within the Elliott Bay samples: one Seattle waterfront station (NMFS #10015) grouped separately from the three Duwamish River stations.

The above analyses were based on benthic samples collected and analyzed by different investigators. In the case of the NMFS samples, benthic samples were collected seasonally for analysis, while sediment chemistry data were available only for one occasion. The same sediment samples were not analyzed for benthic species and chemistry. Differences in parameters measured for different data sets, detection limit variations, and other factors reflecting different methodologies and investigators precluded full use of the data. A similar data set collected as part of a single coordinated study, would provide much more definitive information than that obtained herein. Such a study design should analyze the same sediments for benthos, chemistry and bioassay, as recommended by Chapman and Long (1983) to eliminate concern related to small scale variations in the sediments (cf. Swartz et al., 1982). In addition, a higher number of reference samples is recommended; a ratio of 25% reference stations compared to 75% study stations would greatly improve the statistical confidence of the analyses.

As a result of the analyses undertaken herein, it is recommended that the Case Inlet station (NMFS #12062) not be used in future as a reference area. Although Case Inlet was less degraded than stations from more urbanized embayments, it is more similar in these analyses to the urban stations than to Samish Bay. This conclusion is supported by data from Table 7, which indicated that Case Inlet had a low proportion of arthropod species, a group considered to be relatively sensitive to pollution (Caracciolo and Steimle, 1983). Case Inlet sampling was conducted in an area documented in the 1960's as comprising anaerobic sediments, although the reason(s) for this condition were not determined (J. Word, pers. comm.).

4.4 Correspondences Between Sediment Chemistry, Benthic Communities and Sediment Bioassays

A final step in analysis of the available data involved correlations between sediment chemistry, benthic communities and sediment bioassays, the "Triad" Index (Chapman and Long, 1983). Such analyses could not be done using statistical techniques because of the data inadequacies in the parameters measured (chemistry and bioassay), and methodological differences in the measurements undertaken. Consequently, this comparison was made in a less rigorous fashion.

A graphical presentation of the Triad comparison is shown in Figures 6 and 7 for all stations. Data on benthic communities were extracted from Tables 7 and 8. Data on the concentrations of metals and selected organics in sediments, as available, are provided in Table 13. A sediment bioassay summary index was developed as shown in Table 14, using all available sediment bioassay data. This included bioassay data gathered in different studies for samples taken from approximately the same areas, and hence encompassed a wider data base than the overlapping bioassay data shown in Tables 1, 2, and 3.

Figure 5 clearly shows that the reference areas (Case Inlet, Samish Bay) have a lower level of pollution-related parameters than do urbanized Elliott Bay, Commencement Bay Waterways and Sinclair Inlet. Figures 6 and 7 support this trend, although the Denny Way CSO stations are notable for the presence of a few sensitive phoxocephalid amphipods, and Commencement Bay Waterway stations L1, A6 and S1 include only one type of sediment bioassay test.

On the basis of the Triad data, all stations from the urban areas are degraded compared to the reference stations (higher chemical levels, sensitive amphipods generally absent). This conclusion is in accord with more detailed analysis of the benthic data alone, and with analysis of the benthic and chemistry data together. Although the Triad comparison, at the present level of detail, does not permit differentiation of specific sub-areas based on particular chemicals and taxa, it consistently defines polluted stations. There is, moreover, generally good agreement between the three components of the Triad: benthic community structure, sediment chemistry and sediment bioassays. These results recommend the use of the Triad in future marine pollution assessments as all three components are essential for comprehensive assessment. Individually, chemical analyses provide no information on availability/toxicity to resident biota; community structure does not provide sufficient resolution of contaminated areas and may be affected by factors other than contaminant effects; and, bioassays provide direct evidence of contaminant availability and of adverse contaminant effects but under laboratory conditions not necessarily reflective of real-world environments.

This study represents the first attempt to apply the Triad approach and a number of refinements in the Triad in Figures 6 and 7 should be possible. For instance, a variety of benthic community indices other than those used herein exist (cf. Washington, 1984) and may be applicable. The present simple benthic indices do, however, appear to be more applicable than the Infaunal Trophic Index (ITI) results

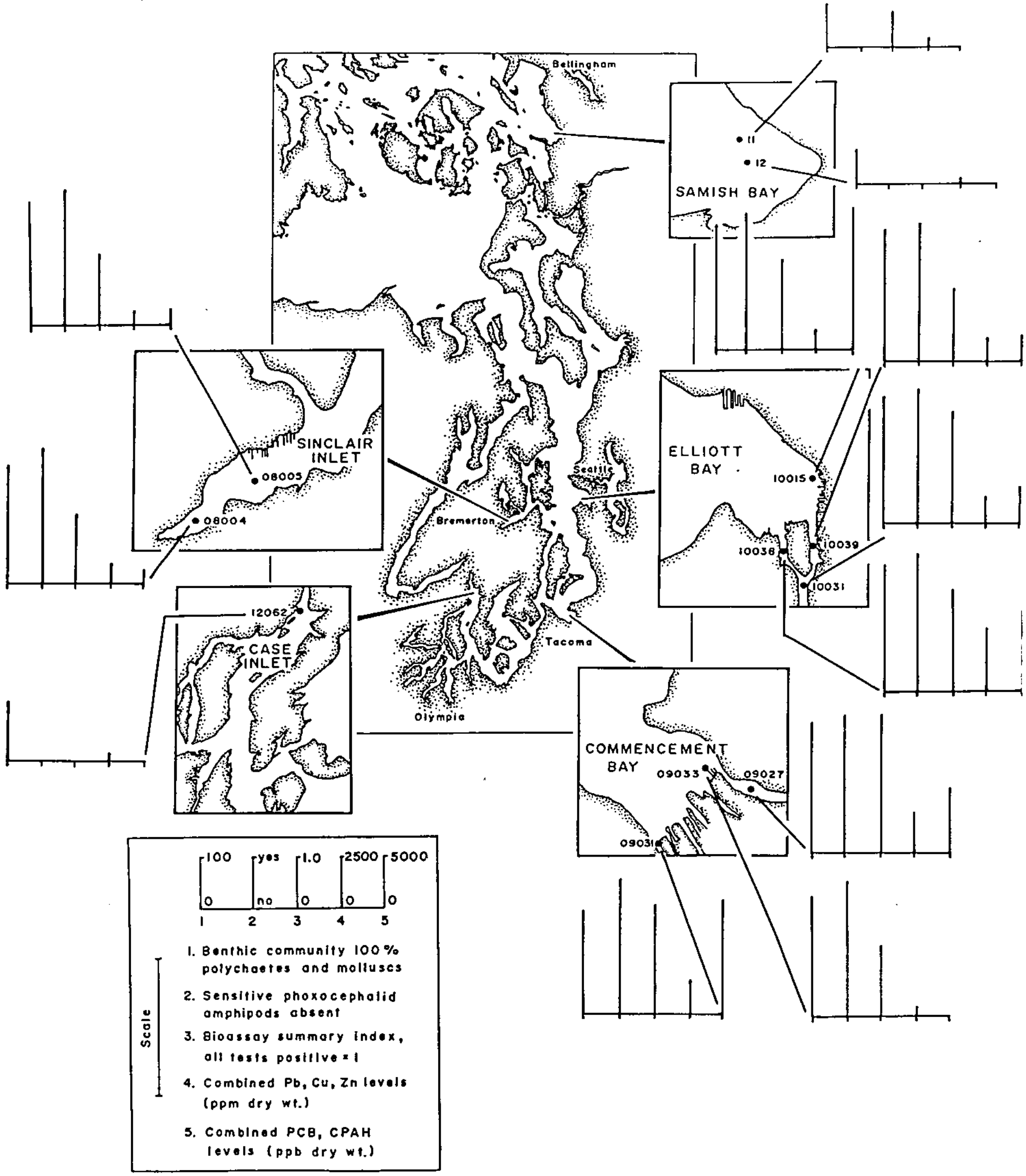

Figure 6. Bar diagrams of Triad sediment data for NMFS samples. Triad = sediment chemistry, sediment bioassay, infaunal benthos.

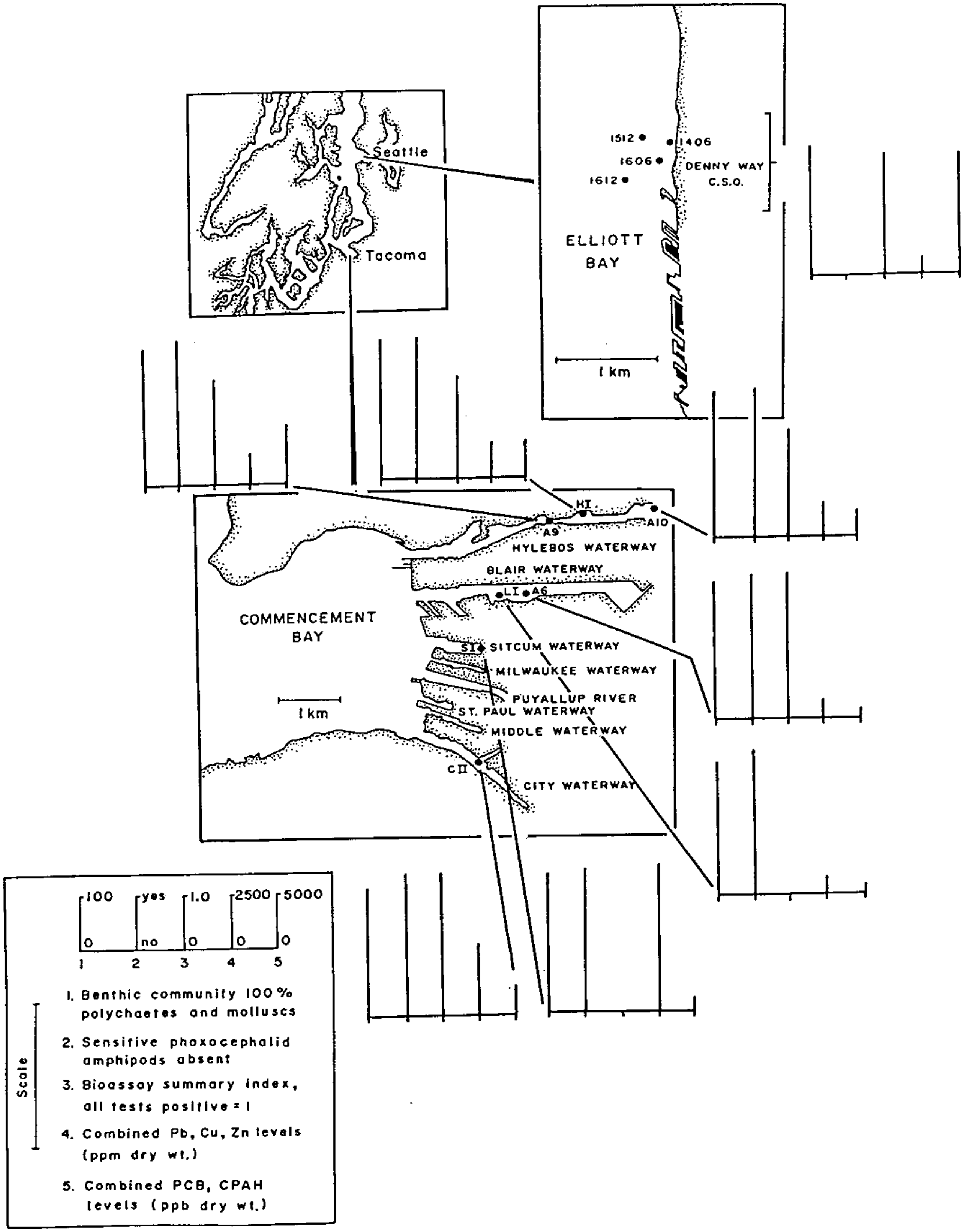

Figure 7. Bar diagrams of Triad sediment data for EPA (Commencement Bay Waterways) and Metro Seattle (Elliott Bay) samples.
Triad = sediment chemistry, sediment bioassay, infaunal benthos.

Table 13. Summary and synthesis of sediment chemistry data for all stations. Data from Malins et al. (1980, 1982), Schults et al. (unpublished report) and EPA Region X (unpublished data).

		METALS[a]				ORGANICS		
Area	Station	Pb	Cu	Zn	Σ Pb, Cu, Zn	PCB[b]	CPAH[c]	$\bar{O}$[d] = (ΣPCB + CPAH/10)
Samish Bay	11	10	15	41	66	N.D.[e]	N.D.	N.D.
	12	46	42	93	181	N.D.	N.D.	N.D.
Case Inlet	12062	24	45	83	152	4	480	52
Elliott Bay	10015	111	91	133	335	492	46500	5142
	10038	627	206	319	1152	665	25000	3165
	10031	265	131	204	600	533	7710	1304
	10039	160	109	175	444	338	3880	726
	1406	89, 2109	130, 87	140, 170	413	1930	34520[f]	5392
	1512	54, 94	52, 55	100, 110	233 $\bar{x}$=286	1416	19947	3411 $\bar{x}$ = 4104
	1606	88, 49	57, 34	150, 86	232	2823	4945	3318
	1612	90, 98	62, 54	120, 110	267	1112	31938	4306
Commencement Bay	09027	154	259	324	737	1150	12530	2403
	09031	269	178	224	671	383	38830	4266
	09033	28	51	57	136	N.D.	500	50
	A9	147	179	202	528	1329	3850[h] (6417)	1971
	A10	123	173	259	555	Trace	4871[h] (8118)	812
	HI	197	211	334	742	390	6120[h] (10200)	1410
	LI	74	106	132	312	N.D.	1442[h] (2403)	240
	A6	69	72	132	273	Trace	1055[h] (1758)	176
	SI	791	581	1190	2562	N.D.	2565[h] (4275)	428
	CII	225	276	742	1243	Trace	7005[h] (11675)	1168
Sinclair Inlet	08004	98	151	156	405	176	2690	445
	08005	136	184	238	558	218	1960	414

a. ppm dry weight.

b. ppb dry weight, Σ 1242, 1254, 1260 or Σ 2-CB, 3-CB, 4-CB, 5-CB, 6-CB, 7-CB, 8-CB, 9-CB, 10-CB.

c. ppb dry weight, = Σ of fluoranthene, pyrene, benz(a)anthracene, chrysene, benzofluoranthenes, benzo(e)pyrene, perylene, indeno(1,2,3-CD)pyrene.

d. Organic average = ΣPCB + ΣCPAH (combustion PAH)/10; division by 10 reduces the CPAH values to approximately equal weight in the sum as the PCBs.

e. N.D. = no data or below detection limits.

f. Perylene was not reported; since perylene is normally less than 10% of the Σ CPAH, no correction was made.

g. Two values reported; averaged to form the sum, Σ Pb, Cu and Zn.

h. The CPAHs for these samples only included fluoranthene, pyrene, benzo(a)anthracene and chrysene; based on the average contribution of these four to the other Σ CPAH values in this table (60%), a comparable Σ CPAH value was calculated and used for the summation.

Σ = sum

Table 14. Summary and synthesis of sediment bioassay data for all stations[a]

Area	Station	Bioassay Data[b]: Amphipod Lethality	Oligochaete Respiration	Oyster Larvae Abnormality	Fish Cell Effects	Polychaete Life-Cycle Effects	Surf Smelt Partial Life-Cycle Effects	Summary Index[c]
Samish Bay	11	no	no	no	yes	N.D.	N.D.	0.25
	12	no	no	no	no	N.D.	N.D.	0.0
Case Inlet	12062	no	N.D.	N.D.	N.D.	N.D.	N.D.	0.0*
Elliott Bay	10015	yes	yes	no	yes	no	yes	0.67
	10038	yes	yes	N.D.	no	N.D.	N.D.	0.67
	10031	yes	yes	yes	yes	no	yes	0.83
	10039	N.D.	no	N.D.	yes	N.D.	N.D.	0.50
	1406 1512 1606 1612	yes	yes	N.D.	yes	no	yes	0.80[d]
Commencement Bay	09027	N.D.	yes	N.D.	yes	N.D.	N.D.	1.0
	09031	N.D.	yes	no	yes	yes	yes	0.80
	09033	N.D.	no	N.D.	yes	N.D.	N.D.	0.50
	A9	no	yes	N.D.	yes	N.D.	N.D.	0.67
	H1	yes	yes	N.D.	yes	N.D.	N.D.	1.0
	A10	no	yes	N.D.	yes	N.D.	N.D.	0.67
	L1	no	N.D.	N.D.	N.D.	N.D.	N.D.	0.0*
	A6	yes	N.D.	N.D.	N.D.	N.D.	N.D.	1.0*
	S1	no	N.D.	N.D.	N.D.	N.D.	N.D.	0.0*
	C11	yes	yes	N.D.	yes	N.D.	N.D.	1.0
Sinclair Inlet	08004	N.D.	no	N.D.	yes	N.D.	N.D.	0.50
	08005	N.D.	yes	N.D.	no	N.D.	N.D.	0.50

a. Data were extracted, as appropriate, from the following sources: Chapman et al., 1982, 1983, 1984; Chapman and Fink, 1983; Ott et al., 1983; Swartz et al., 1982.

b. Yes = significant (positive) result; no = non-significant (negative) result; N.D. = no data.

c. Index values are calculated as the total number of positives over the total number of positive and negative results.

d. These stations are considered as a group due to their location in close proximity to each other.

*Indicates that values are based on only one measured parameter, and require verification by one or more different bioassay tests.

reported by Malins et al. (1982a). These authors reported ITI values for the NMFS stations analyzed herein (excepting NMFS #09031) as 60 or greater, that is, indicating no organic enrichment. Malins et al. (1982a) also used a form of diversity index, noting that the greatest number of species (=taxa as these authors did not undertake species-level identification) occurred in reference areas, with lowest species numbers in urban waterways.

The bioassay summary index used herein was based on the available data which included a variety of different, non-overlapping tests. The conduct of the same tests at all sites would provide better data for quantification of this parameter. Similarly, chemistry data used for the Triad could be improved.

The fact that the present, preliminary Triad approach provides useful, verifiable data on areas of degradation supports further studies using this approach. Such studies should include development of final indices for Triad parameters.

4.5 Areas of Significant Degradation In Puget Sound

Separate measures were used to determine areas of significant degradation in Puget Sound, based on the available data. Forensic, but non-statistical analysis of the benthic community data (Section 4.1) indicated that samples from the urban embayments tested were degraded compared to those from the reference areas, based on the presence of high proportions of the total infauna represented by polychaetes and molluscs at these stations, and the general absence of more sensitive species such as echinoderms and arthropods.

The use of the IBD also indicated that the study areas were significantly degraded compared to the reference areas. Index values were substantially greater than those described by O'Connor (unpublished report) as indicative of marginal degradation. Calculation of the IBD for Commencement Bay Waterways and Elliott Bay separately indicated a slightly higher level of degradation for Commencement Bay Waterways.

Multivariate analyses of the benthic data supported the above analyses but indicated that the Case Inlet station was more similar to the urban embayments than the Samish Bay stations. Whether these results are reflective of temporal differences (Case Inlet samples were collected in 1979 whereas Samish Bay samples were collected in 1983) or spatial differences (Samish Bay is located north of Admiralty Inlet), is unknown. Indications that the various urban embayments were further separable could not be confirmed with benthic data alone.

Multivariate analyses using combined benthos and chemistry data indicated that Case Inlet was more similar to the urban embayments, in particular Sinclair Inlet, than Samish Bay. Urban embayments were separable based on a combination of factors including specific species distributions and PAH concentrations, however this separation could not describe a measure of relative degradation by area.

Implementation of the Triad approach of Chapman and Long (1983) confirmed the degraded/polluted status of the urban embayments compared to the reference areas. This approach also provided indications that individual stations (representing specific sub-areas within urban embayments) might be separable with refinement of this technique.

On the basis of the above analyses, the three urban embayments (Elliott Bay, Sinclair Inlet, Commencement Bay Waterways) are areas of significant degradation in Puget Sound. Case Inlet appears to be less useful as a reference area than was initially suspected, while Samish Bay appears to be the most suitable reference area of those tested.

Elliott Bay may be less degraded than Commencement Bay Waterways based on the comparative IBD calculation but sediment contaminant levels are generally higher in Elliott Bay than in Commencement Bay (Quinlan et al., in press) and more taxa had reduced abundances in Elliott Bay as compared to Commencement Bay Waterways. The results of the multivariate analyses suggest that Sinclair Inlet is less degraded than Elliott Bay and Commencement Bay Waterways. However, the small number of stations examined in this study precludes a valid prioritization of areas by level of degradation.

5.0 CONCLUSIONS

The following major conclusions can be derived from the results of this study, related to the study objectives:

1. Data collected by NMFS, EPA and Metro on benthic communities in Puget Sound provided useful comparative information even though different techniques and investigators had been involved in collections and identifications.

2. Degraded areas in Puget Sound are generally characterized by a high proportion of polychaetes and molluscs, a low proportion of arthropods and echinoderms, and a general absence of sensitive amphipod families (Phoxocephalidae, Lysianassidae).

3. Stations and areas of Puget Sound with higher levels of chemical contamination and sediment toxicity contain benthic infaunal communities indicative of environmental degradation.

4. Elevated sediment contaminant levels and positive toxicity test results correspond with and are possibly indicative of actual effects on benthic communities.

5. The Index of Benthic Degradation (IBD), although designed to determine areas of marginal degradation (its original application), is a functional tool for comparing degraded areas (application used in the present study).

6. The Triad approach, combining analyses of sediment chemistry, sediment bioassays, and benthic infaunal distributions, provides additional resolution in determining and prioritizing areas of degradation. Information is provided not only on areas of contamination, but also on associated sediment toxicity and changes to communities of resident benthic biota.

7. Both the IBD and the Triad approaches are in the developmental stage. However, since the Triad approach incorporates three environmental components while the IBD incorporates only one component, the Triad approach utilizes more independent observations related to environmental degradation.

8. All three urban embayments studied are significantly degraded based on selected samples. In comparative terms, Elliott Bay may be slightly less degraded than Commencement Bay Waterways, with Sinclair Inlet exhibiting the least degradation.

9. There is evidence of environmental degradation in Case Inlet (a reference area), although this area is less degraded that the three urban embayments. Samish Bay provided a better reference area (less obviously affected by pollution).

6.0 RECOMMENDATIONS

The following major recommendations result from the present study.

1. Species-level taxonomic analyses of archived NMFS benthos samples yielded useful information in the present study. However, only a portion of these samples have been analyzed. We recommend that all of the remaining archived NMFS samples be subjected to species-level taxonomy and that this data be available for ongoing assessment in Puget Sound.

2. Use of the EPA, NMFS and Metro data sets required standardization of species names. To improve the usefulness of future benthic studies in Puget Sound, a program of taxonomic standardization should be implemented including exchange of information and specimens between various investigators.

3. The Triad approach is a useful and informative method of determining areas of environmental degradation and of relating laboratory studies to in situ situations. We recommend that this approach be verified using a better data set than that available for the present study. Specifically, all data for verification of the Triad should be collected as part of a single, separate, synoptic integrated study. Such a study should examine areas of low, medium and high environmental degradation, and 25% of the stations tested should be from reference areas.

4. Indices (=measures of effects/differences) for use in the three component parts of the Triad should be better defined. In this connection the IBD may provide a useful measure of benthic community effects.

5. The Triad should be adopted by NOAA as one of its forthcoming "Indices of Environmental Degradation". Unlike other indices presently under consideration, the Triad addresses sediment toxicity directly.

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