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# Progress Re-evaluating Puget Sound Apparent Effects Thresholds (AETs)

Volume I:  
1994 Amphipod and Echinoderm Larval AETs

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For  
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DRAFT Report  
April 1996

## Acknowledgements

The analysis and discussion in this document were prepared by Thomas H. Gries and Kathryn H. Waldow. Preliminary review was provided by staff of Ecology's Sediment Management Team, chiefly Brett Betts, Rachel Friedman-Thomas, Maria Peeler and Keith Phillips. The following Puget Sound Dredged Disposal Analysis (PSDDA) program staff provided additional technical review:

U.S. Army Corps of Engineers, Seattle District  
David Fox, David Kendall, Therese Littleton, Stephanie Stirling  
U.S. Environmental Protection Agency, Region 10  
John Malek, Justine Barton  
Washington Department of Natural Resources  
Phil Hertzog, Deborah Lester, Gene Revelas

In addition to those mentioned above, the authors are grateful to

- the numerous people, from various agencies and groups, who provided sediment quality data and quality assurance information to update Ecology's sediment quality values database
- Kathy Bragdon-Cook, A. Martin Payne, Desiree Brown-Turner, and Tuan Vu of Ecology for their cheerful perseverance in reviewing and entering many of those data
- Nigel Blakely, James Cabbage, Bill Eyinger, Lon Kissinger of Ecology and Travis Shaw (Corps) for their statistical expertise
- Bob Barrick, Scott Becker, Dreas Nielsen, Rob Pastorok of PTI Environmental Services, Inc., for sharing undocumented knowledge of the 1988 sediment quality values refinement process, valuable advice and essential software support
- Ecology and the PSDDA agencies for their direction and abundant patience
- the many consultants and scientists who contributed their comments and feedback throughout this process, especially in response to later drafts of this document

# TABLE OF CONTENTS

LIST OF TABLES .....	iii
LIST OF FIGURES .....	iv
LIST OF ACRONYMS .....	v
EXECUTIVE SUMMARY .....	vii
INTRODUCTION .....	1
BACKGROUND .....	3
Apparent Effects Thresholds (AETs) .....	3
Puget Sound Dredged Disposal Analysis (PSDDA) .....	3
Sediment Management Standards (SMS) .....	6
Goals and Objectives .....	7
METHODS .....	9
General Approach .....	9
Data Acquisition .....	9
Chemistry: Analytical Methods and Data Quality Assurance .....	11
Bioassays: Analytical Methods and Data Quality Assurance .....	12
Bioassays: Performance of Control and Reference Samples .....	13
Determination of Adverse Biological Effects .....	14
Significant Amphipod Mortality .....	15
Significant Sediment Larval Effects .....	16
Chemically Anomalous Samples .....	17
Calculating AETs and Predictive Reliability .....	18
RESULTS .....	23
Inventory of Synoptic Data for Puget Sound .....	23
Data Excluded from AET Calculations .....	23
The Puget Sound AET Database: 1988 and 1994 .....	24
Dry Weight-Normalized Values and Samples Setting Them .....	25
.....	27
TOC-Normalized Values and Samples Setting Them .....	27
Overview of Sediment Larval AETs .....	28
Echinoderm Larval Abnormality AETs .....	29

Dry Weight-Normalized Values and Samples Setting Them .....	29
TOC-Normalized Values and Samples Setting Them .....	29
Predictive Reliability of 1994 AETs .....	30
Amphipod Mortality AETs .....	30
Echinoderm Larval Abnormality AETs .....	47
Some Characteristics of Incorrectly Predicted Stations .....	47
Reliability of Pooled AETs .....	53
CONCLUSIONS AND DISCUSSION .....	55
The 1994 AET Database .....	55
Methods .....	55
1994 Puget Sound AET Values .....	56
New HAETs, 2AETs, LAETs: Potential Guidelines and Standards .....	58
Reliability of 1994 Puget Sound AETs .....	66
Summary .....	69
RECOMMENDATIONS FOR FUTURE WORK .....	71
REFERENCES .....	73
GLOSSARY .....	75
APPENDIX A: Methods .....	A-1
APPENDIX B: The 1994 Sediment Quality Values Database (SEDQUAL) .....	B-1
APPENDIX C: 1994 Puget Sound AET and Reliability Results (on diskette) .....	C-1

## LIST OF TABLES

- Table 1. Bioassay sample performance standards.
- Table 2. Comparison of 1988 and 1994 Puget Sound AET databases.
- Table 3. 1994 amphipod mortality AETs, dry weight-normalized.
- Table 4. 1994 amphipod mortality AETs, TOC-normalized.
- Table 5. 1994 echinoderm larval abnormality AETs, dry weight-normalized.
- Table 6. 1994 echinoderm larval abnormality AETs, TOC-normalized.
- Table 7. Summary of predictive reliability of 1994 AETs.
- Table 8. Summary of characteristics of "Hit" samples incorrectly predicted by 1994 AET values.
- Table 9. Suite of five dry weight-normalized AETs for Puget Sound.
- Table 10. Suite of five TOC-normalized AETs for Puget Sound.
- Table 11. Summary of 1994 highest, second lowest and lowest AET values.

## LIST OF FIGURES

- Figure 1. Determination of an Apparent Effects Threshold (AET) value.
- Figure 2. General approach used to calculate 1994 Puget Sound AETs.
- Figure 3. Predictive reliability of AET values.

## LIST OF ACRONYMS

2AET	-	The second (2nd) Highest Apparent Effects Threshold
AET	-	Apparent Effects Threshold
AMPT	-	AMPhipod Toxicity bioassay
ARM	-	PSDDA Annual Review Meeting
BENA	-	BENThic Abundance
CSL	-	Cleanup Screening Level
DDD	-	DichloroDiphenylDichloroethane
DDE	-	DichloroDiphenyldichloroEthylene
DDT	-	DichloroDiphenylTrichloroethane
DO	-	Dissolved Oxygen
DNR	-	Washington State Department of Natural Resources
EC <sub>50</sub>	-	Effective Concentration, 50%
ECHN	-	ECHiNoderm larval toxicity bioassay
EPA	-	U.S. Environmental Protection Agency
HAET	-	Highest Apparent Effects Threshold
HPAH	-	High molecular weight Polynuclear Aromatic Hydrocarbon compounds
LAET	-	Lowest Apparent Effects Threshold
LC <sub>50</sub>	-	Lethal Concentration, 50%
LPAH	-	Low molecular weight Polynuclear Aromatic Hydrocarbon compounds
MCUL	-	Minimum CleanUp Level
MICB	-	MICrotox <sup>®</sup> luminosity Bioassay (saline extract)
ML	-	PSDDA program Maximum Level guideline concentrations
NMFS	-	National Marine Fisheries Service
NOAA	-	National Oceanic and Atmospheric Administration
NPDES	-	National Pollutant Discharge Elimination System
OYST	-	OYSer larval Toxicity bioassay
PAH	-	Polynuclear Aromatic Hydrocarbon compounds
PCB	-	PolyChlorinated Biphenyls
PSEP	-	The Puget Sound Estuary Program
PSDDA	-	The Puget Sound Dredged Disposal Analysis agencies and program
PSWQA	-	Puget Sound Water Quality Authority
QA	-	Quality Assurance.
QA1	-	Quality Assurance, Level 1.
QA2	-	Quality Assurance, Level 2..

SEDQUAL	-	Ecology's <b>SED</b> iment <b>QUAL</b> ity values or AET database.
SIZ <sub>max</sub>	-	The <b>maximum</b> allowable Sediment Impact Zone
SL	-	PSDDA program Screening Level guideline concentration.
SMARM	-	Sediment Management Annual Review Meeting
SMS	-	Washington State Sediment Management Standards.
SRM	-	Standard Reference Material
SQS	-	Sediment Quality Standard
SQV	-	Sediment Quality Value
TBT	-	TriButyl Tin
TOC	-	Total Organic Carbon.
TVS	-	Total Volatile Solids
USACE	-	U.S. Army Corps of Engineers

## EXECUTIVE SUMMARY

Apparent Effects Threshold (AET) values are the concentrations of specific chemicals of concern (COC) in sediment above which a significant adverse biological effect or "Hit" always occurs. They form the basis for both the Puget Sound Dredged Disposal Analysis (PSDDA) program guidelines (1,2) and the criteria contained in the Sediment Management Standards (SMS) rule (3).

This document is the result of the first extensive re-evaluation of sediment AETs since 1988. The re-evaluation uses extensive new synoptic data to 1) recalculate amphipod mortality AETs, 2) calculate new sediment larval AETs, and 3) determine the predictive reliability of both AET groups. A further key objective is to assess some of the potential implications of new AETs for the PSDDA and SMS programs, as well as the dredging community and other stakeholders. The scope of the re-evaluation does not include calculating AETs based on new benthic infaunal, saline extract Microtox® or 20-day juvenile polychaete (*Neanthes arenaceodentata*) endpoints.

PSDDA program staff compiled an extensive inventory of matching chemistry and bioassay, or "synoptic," samples collected up until March 1993. Many of these samples, however, were excluded from final AET calculations because they failed to meet chemical quality assurance (QA) requirements or else lacked an adequate negative control or reference area sample. Even with data exclusions, the 1994 AET re-evaluation began with a sediment quality values database that contained more than double the 1988 number of amphipod mortality samples and over 200 echinoderm larval samples.

The PSDDA agencies attempted to adhere closely to the quality assurance, biological effects interpretation, AET and reliability calculation methods used in 1988 (4). This was done to avoid having to repeat certain discussions or controversies. The report does describe some method variations, though. For example, 1994 AET calculations included subsurface synoptic samples which met all general quality assurance requirements. Some of the newer sample chemistry data have yet to be fully quality assured to meet the more stringent "QA2" level requirements, as was done in 1988. Sediment larval bioassay results were interpreted using the abnormality endpoint alone and also effective mortality (abnormality plus mortality). Statistical comparisons made between test and reference samples differed slightly from those used in 1988. However, none of these was thought to have substantially affected the final 1994 AET or reliability values.

Most of the amphipod mortality AETs calculated in 1994 either remained the same or increased relative to the corresponding 1988 values, whether normalized to dry weight or organic carbon. AETs for trace metals and individual HPAHs were most affected. Dry weight-normalized arsenic, cadmium, lead, mercury and zinc AET values increased by factors ranging from 1.1 to 4.8. Six individual dry weight HPAH AETs increased by an average factor of 2.2.

The sensitivity measure of reliability for dry weight-normalized amphipod AETs declined from 58% in 1988 to 43% in 1994. However, the overall reliability of 1994 values was similar to that found in 1988, 84% and 85%, respectively.

In early 1993, the number of newer bivalve larval bioassay samples was not adequate to calculate separate bivalve larval AETs. Therefore, and on the advice of marine benthic experts, echinoderm data alone were used to calculate separate, new AET groups. These echinoderm larval AETs were based on both the abnormality endpoint and the effective mortality (abnormality + mortality) endpoint. AET values derived using the former endpoint proved more sensitive and reliable overall, so the final 1994 echinoderm larval AETs presented in this report were calculated using only abnormality.

Twenty-seven of the dry weight-normalized echinoderm abnormality AET values calculated in 1994 were lower and seven were higher than corresponding 1986 oyster values. Overall, TOC-normalized echinoderm AETs were also lower than oyster AETs. These lower echinoderm AETs may indicate a fundamentally lower tolerance of the bioassay test organisms, primarily *Dendraster excentricus*, toward chemical contaminants in Puget Sound sediments.

Reliability calculations revealed that the 1994 dry weight-normalized echinoderm abnormality AETs were far less sensitive than 1986 oyster AETs: 48% vs. 88%. However, the independent reliability calculations indicated they were substantially more efficient and had better overall reliability than the oyster AETs. Echinoderm AETs were the second most efficient of all AET types. Sensitivity and overall reliability for TOC-normalized echinoderm abnormality AETs were both reduced relative to 1986 oyster values.

Both 1994 amphipod mortality and echinoderm abnormality AETs were examined as part of a new suite of five possible AETs for Puget Sound: 1994 amphipod, 1988 benthic, 1994 echinoderm, 1986 Microtox and 1986 oyster AETs. The highest dry weight-normalized AET (HAET) values for 17 COCs were greater than reported in 1988. Most of the new HAETs were for trace metal or HPAH compounds, and were set by the recalculated 1994 amphipod AETs. Echinoderm

AETs, particularly dry weight-normalized ones, set numerous new second lowest AETs (2AET) and lowest AETs (LAET).

New HAETs set by dry weight-normalized amphipod mortality AET values have potential implications to the PSDDA program for revising ML and SL guidelines. Echinoderm AETs, which much more frequently established new 2AET or LAET values, have potential to change the SMS criteria and program.

However, proposing changes to the guidelines or the criteria used in either program will involve detailed review of the AET reliability results presented in this report, as well as other reliability calculations yet to be conducted. The values eventually adopted by either the PSDDA or SMS program will likely not compromise certain reliability measures of the current regulatory values. Finally, the practical and economic implications of any changes to PSDDA MLs and SLs, and SMS criteria, will also need to be considered prior to adopting new regulatory values.

## INTRODUCTION

Management of dredged material and contaminated sediment in Washington State is based on numerical sediment quality values. Two examples of these are the PSDDA program guidelines (1,2) and the criteria contained in the SMS rule (3). Both are derived from AET values which were last calculated in 1988 using data collected from throughout Puget Sound between March 1982 and September 1986 (4).

This report represents the first extensive re-evaluation of sediment AETs since 1988. It is driven in part by the availability of additional sediment quality data, many of which are incorporated into recalculating AETs and their reliability. Results include a list of revised amphipod mortality AETs, new echinoderm larval AETs, and a comparison of these "1994" AETs to 1986 and 1988 values. These are presented and discussed in a PSDDA program context, but may also facilitate review of the SMS rule.

By re-evaluating AETs and their potential to affect the PSDDA maximum and screening levels (MLs and SLs), the report addresses an agreement to conduct an annual review of the dredging program (2). It also partly fulfills Ecology's requirement to annually review the SMS rule, and the source control and cleanup criteria contained therein.

While this is a technical report, it is intended for experts and the public alike. It presents methods and results in a manner that can be repeated by future investigators, but understood by those less familiar with sediment management in Puget Sound. It offers some objective conclusions and discussion for both audiences. The report does not, however, suggest policy options or recommend specific actions relative to the use of new AET values. Regulators, in particular, should make note of this.

The report is organized into five main sections. A **Background** section provides basic information on AETs and how they are used in Puget Sound. It also outlines goals and objectives of the report. The **Methods** section details how 1994 AETs were re-calculated. **Results** contains new amphipod mortality and echinoderm larval AET values, as well as their ability predict adverse biological effects. The section which follows offers **Conclusions and Discussion**. Finally, a brief **Recommendations for Future Work** lists important next steps and suggests some possible refinements to re-evaluation methods.

In addition to the main sections, the reader will find the **Table of Contents** followed by a **List of Acronyms** (pages v-vi) and an **Executive Summary** (pages vii-ix). A **Glossary** of terms located immediately after the **References** section.

References are cited throughout the text as italicized numbers within parentheses, e.g., (3).

Three separate appendices accompany this report. **Appendix A** details the methods used to conduct bioassays, interpret biological effects, calculate AETs and reliability. **Appendix B** characterizes the sediment quality values database used to calculate 1994 AETs. The final appendix, **Appendix C**, includes more complete results of AET and reliability calculations. It is provided on a single 3.5" floppy diskette as a series of spreadsheet files (Excel, version 4.0).

Finally, it is important to note that Volume I of this report does not assess the potential implications that new AETs may have on regulatory programs. That will be done under the direction of a "Regulatory Work Group" or other team of experts and will be presented in a second volume.

## BACKGROUND

### Apparent Effects Thresholds (AETs)

*AETs are concentrations of specific chemicals of concern (COC) in sediment above which a significant adverse biological effect always occurs.* They are empirically derived using synoptic sediment samples - those having undergone simultaneous testing for chemical contaminants and adverse biological effects.

Determination of a single AET value is depicted in Figure 1. All synoptic samples that do not exhibit significant adverse effects, "No Hit" samples, are ranked from highest to lowest concentration for a chemical of concern. The "No Hit" sample with the greatest concentration is identified and generally establishes the AET value. A rare exception is made when that sample is found to be chemically anomalous (see Methods and Glossary). In that case, the sample with the next highest concentration sets the AET.

There must be at least one sample exhibiting a significant adverse effect, i.e., one "Hit" sample, with a chemical concentration exceeding the AET to confirm that AET value. If no "Hit" sample has a greater concentration, then the AET is qualified as a minimum value with a "G" or ">" symbol.

The determination of a single AET is repeated for all chemicals of concern. The resulting group of AETs is for a single biological indicator, interpretive endpoint, unit of measure and measurement basis. For example, one may calculate dry weight-normalized amphipod mortality AETs and express them in units of ppm. However, the determination process can be repeated to generate AET groups based on other biological indicators, endpoints, measurement units or bases. The 1988 AETs include values based on amphipod mortality, benthic infaunal abundance, Microtox<sup>®</sup> luminosity and oyster larval abnormality.

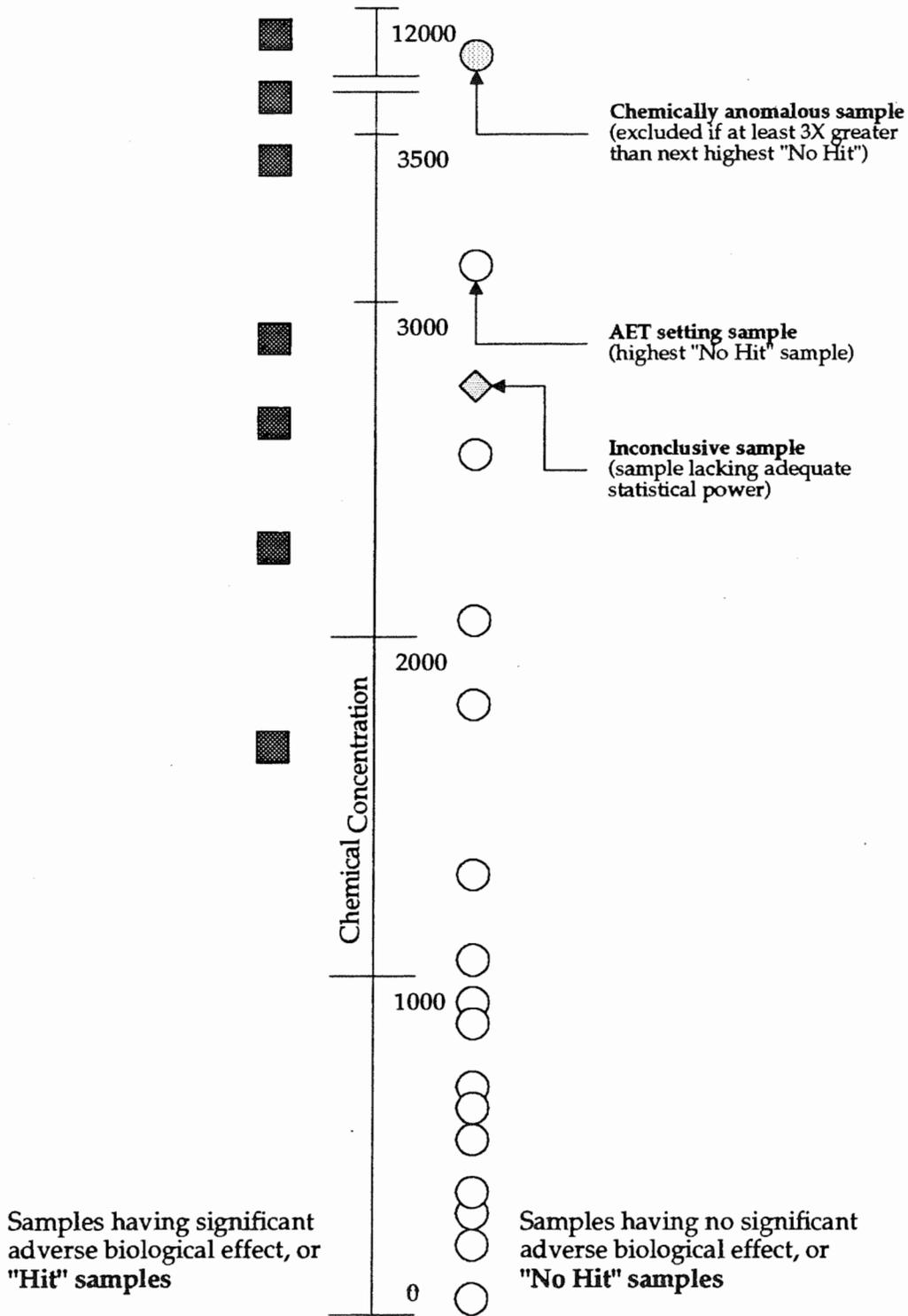
As stated in the Introduction, AET values are used by both the PSDDA and SMS programs to establish numerical guidelines and criteria. The relationship between the various AET and regulatory values, together with how the latter are used, is described below.

### Puget Sound Dredged Disposal Analysis (PSDDA)

The PSDDA program routinely uses MLs and SLs to make decisions on the management and open water disposal of material to be dredged from Puget Sound. Current MLs and SLs are mainly based on 1988 AETs for the four biological indicators of toxicity just listed (4). Those AETs incorporate extensive sediment quality data sets from Eagle Harbor, Elliott Bay and Everett Harbor

**Figure 1. Determination of an Apparent Effects Threshold (AET) value.**

An AET is generally set by the sample with the highest chemical concentration of a potential toxicant which does not exhibit a significant adverse biological effect ("No Hit"). The AET is qualified as a minimal value using a "G" or a ">" symbol if no "Hit" sample exceeds it. Note: Any units of measure or means of normalization may apply.



which were not used to determine the original 1986 AETs (5). The highest of the four AET values, or HAET, establishes the ML for a given chemical of concern. The SL is set at one-tenth the ML, provided it is a) less than or equal to the lowest of the four AETs, or LAET, and b) greater than or equal to the average reference area concentration (1,2).

In the PSDDA program, 60 different chemical ML or SL values are used to define three categories of dredged material. Material with COC concentrations exceeding an ML value is generally unsuitable for open water disposal<sup>1</sup> because the ML itself is an indication of several types of significant adverse biological effects. Material having chemical constituents between ML and SL concentrations may be expected to exhibit at least one type of adverse biological effect. Thus, additional biological effects testing is required to provide PSDDA agencies with the information needed for regulatory decisions. Material exhibiting sediment chemistry concentrations below all SL values is considered suitable for open water disposal because none of the adverse biological effects used to establish AETs are expected.

In order for regulatory decisions to protect biological resources, the PSDDA agencies compare high quality sediment data to ML and SL guideline values which reflect adverse biological effects.

One way PSDDA agencies ensure collection of high quality data is by recommending use of certain field and laboratory protocols (6,7). These protocols encompass vessel positioning, collection of sediment samples, chemical analysis, quality assurance review, biological testing and interpretation of biological effects, etc. The PSDDA annual review process and Annual Review Meeting (ARM) facilitate adopting changes to those protocols to reflect "state-of-the-art" science, consensus-based policy making, and public input. The PSDDA process and agency responsibilities are described further in the Phase II Management Plans (1,2, Chapter 9).

But regulatory decisions made by the PSDDA agencies also rely on the best available guidelines. For this reason, the PSDDA Phase II Management Plan requires an annual review of ML and SL values, too. In re-evaluating the guidelines, the agencies may consider and incorporate the following:

- synoptic sediment quality data collected during previous dredging years
- new field and laboratory experiences

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<sup>1</sup> Sediments which exceed two or more ML values, or one ML value by more than 100%, are considered unsuitable for placement at open water disposal sites. Sediments which exceed one ML by less than 100% must undergo biological testing.

- changes to how significant adverse biological effects are defined<sup>2</sup>
- results from new bioassays<sup>3</sup> (10)

Re-evaluations conducted in 1990 and 1991 (8,9) resulted in seven SL values being raised<sup>4</sup>.

In preparation for this re-evaluation of MLs and SLs, PSDDA agencies obtained and reviewed additional synoptic data collected through March 1993. Preliminary AET values for amphipod mortality, bivalve and echinoderm larval abnormality, and combined larval species abnormality were presented at the 1993 and 1994 ARMs (11,12) and elsewhere (13,14). This report incorporates comments on those presentations, as well as subsequent PSDDA agency and peer review feedback.

### Sediment Management Standards (SMS)

In 1991, Ecology adopted 173-204 WAC: the Washington State Sediment Management Standards rule (3). The rule addresses source control and cleanup of contaminated sediments by establishing marine chemical and biological sediment quality standards (SQS), cleanup screening level (CSL), minimum cleanup level (MCUL) and sediment impact zone (SIZ<sub>max</sub>) values. In general, SQS values are based on the LAET, while CSL, MCUL and SIZ<sub>max</sub> values are based on the second lowest of the four 1988 AETs (2AET).

Analogous to the PSDDA program guidelines, 47 chemical standards classify marine sediments according to levels of contamination. Sediments exceeding a CSL value are expected to show some adverse biological effects and may require cleanup<sup>5</sup>. Sediments exceeding a SIZ<sub>max</sub> value may warrant the establishment of a sediment impact zone associated with a discharge permit. Sediments exceeding only SQS values may be expected to show at least one type of biological effect and thus require further characterization. Finally, sediments

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<sup>2</sup> For example, PSDDA agencies adopted minor modifications to the saline extract Microtox<sup>®</sup> protocol and definition of significant adverse effects (2, pages 5-24).

<sup>3</sup> In 1992, PSDDA agencies replaced the 10-day juvenile polychaete mortality bioassay with the 20-day juvenile polychaete growth test. Both use the species *Neanthes arenaceodentata*.

<sup>4</sup> SLs for 1,2,4-trichlorobenzene, 2-methyl-phenol, 2,4-dimethylphenol, benzoic acid, benzyl alcohol, N-nitrosodiphenylamine and pentachlorophenol were increased to 13 ppb, 20 ppb, 29 ppb, 400 ppb, 25 ppb, 28 ppb, and 100 ppb dry weight, respectively.

<sup>5</sup> The SQS concentrations serve as cleanup goals.

with concentrations falling below the SQS are not predicted to exhibit significant adverse biological effects, and thus do not require remediation or creation of an impact zone.

Section 130(6) requires the SMS rule to be periodically reviewed and revised, as necessary (3). According to Section 130(7)(a), "new or additional scientific information which is available relating surface sediment chemical quality to acute or chronic adverse effects on biological resources" must be considered during the SMS review process. As part of the process, any changes to AETs are examined for potential effect on the adopted Puget Sound marine SQS, CSL, MCUL and  $SIZ_{max}$  values<sup>6</sup>.

## Goals and Objectives

The goal of this report is to clearly document the methods and results of the first extensive re-evaluation of AET values since the "1988 Update and Evaluation of Puget Sound AETs" (4).

Specific objectives for this re-evaluation were initially determined by PSDDA agencies and grouped as mandatory, optional, or deferred (10). The consensus among the agencies was that mandatory objectives had to be completed before any change to existing guidelines or criteria could be recommended. Completion of optional objectives was desirable, but was secondary to mandatory ones and dependent on agency resources. Still other objectives had to be deferred until adequate resources or technical guidance became available.

The objectives evolved throughout the project to reflect early analytical results, public comments and variable agency resources. The final objectives are summarized below.

### Mandatory objectives:

- recalculate dry weight-normalized amphipod mortality AETs
- calculate new dry weight-normalized sediment larval AETs
- calculate of Total Organic Carbon or TOC-normalized AETs<sup>7</sup>
- determine the predictive reliability of the new 1994 AET values

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<sup>6</sup> Those chemical concentration criteria are listed in Section 320(2)/Table I, Section 420/Table II, and Section 520(2)/Table III, respectively).

<sup>7</sup> This objective was optional for the PSDDA program but considered mandatory for Ecology's review and revision of the SMS rule.

- determine the combined or "pooled" reliability of the entire new suite of Puget Sound AET values
- assess some of the potential implications of new AETs for PSDDA MLs, SLs, the program as a whole and the dredging community

Optional objectives:

- calculate echinoderm and bivalve larval AET values separately
- compare sediment larval AET values calculated using different bioassay endpoints<sup>8</sup>
- assess some of the potential implications of new AETs for the SMS criteria, the SMS program as a whole and stakeholders

Deferred objectives:

- recalculate of benthic infaunal AETs
- recalculate of saline extract Microtox<sup>®</sup> AETs
- calculate AETs based on the 20-day juvenile polychaete (*Neanthes arenaceodentata*) growth bioassay
- analyze patterns of sediment quality in Puget Sound, as requested by the Washington Public Ports Association (8)

Benthic experts recommended improvements to the interpretive endpoint for benthic infauna sample data (15), but benthic AETs could not be recalculated without final reference sample performance standards. The exercise of recalculating Microtox<sup>®</sup> luminosity AETs awaited additional studies, possibly including side-by-side comparisons to the performance of other bioassays. When this re-evaluation began, the synoptic database for 20-day juvenile polychaete (*Neanthes*) growth bioassays was too limited to calculate AETs<sup>9</sup>. Finally, the pattern analysis mentioned was considered a lower priority than other deferred objectives.

Volume I of this report addresses all but the last of the mandatory objectives. A second volume will describe results of additional reliability analyses, the remaining mandatory objective - assessment of potential implications of new AETs - and a summary of the overall re-evaluation process. Neither volume outlines policy options or recommends actions on the use of new AETs in the PSDDA or SMS programs.

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<sup>8</sup> The PSDDA agencies sought to compare sediment larval AET values based on the abnormality endpoint alone to ones calculated using the combined abnormality + mortality, or "effective mortality," endpoint.

<sup>9</sup> Calculations of AETs result in highly variable values unless the total number of synoptic samples used approaches 50 (4, page 50).

## **METHODS**

### **General Approach**

The general approach used to recalculate AETs is summarized in Figure 2. First, an inventory of synoptic surveys containing bioassay data was compiled. Data for most of those surveys were then obtained and checked for completeness. Complete data sets next underwent a quality assurance (QA) review of chemical and bioassay data. Both types of data had to meet certain minimal guidelines, described later in this section. In addition, bioassay control and reference samples had to meet specific performance standards.

Bioassay test sample results were analyzed for statistically significant adverse effects, relative to one or more reference samples. This was done for all samples meeting the QA guidelines and performance standards. Samples fell into the categories illustrated in Figure 2: those exhibiting significant adverse effects ("Hit" samples), those which did not ("No Hit" samples), and those which were statistically inconclusive. The latter were excluded from calculations, as were "No Hit" samples found to be anomalous. Next, biological effects interpretations - "Hit" and "No Hit" data - were added to Ecology's sediment quality database (SEDQUAL). Finally, the new data were combined with comparable historical data, where possible, and 1994 AETs were calculated.

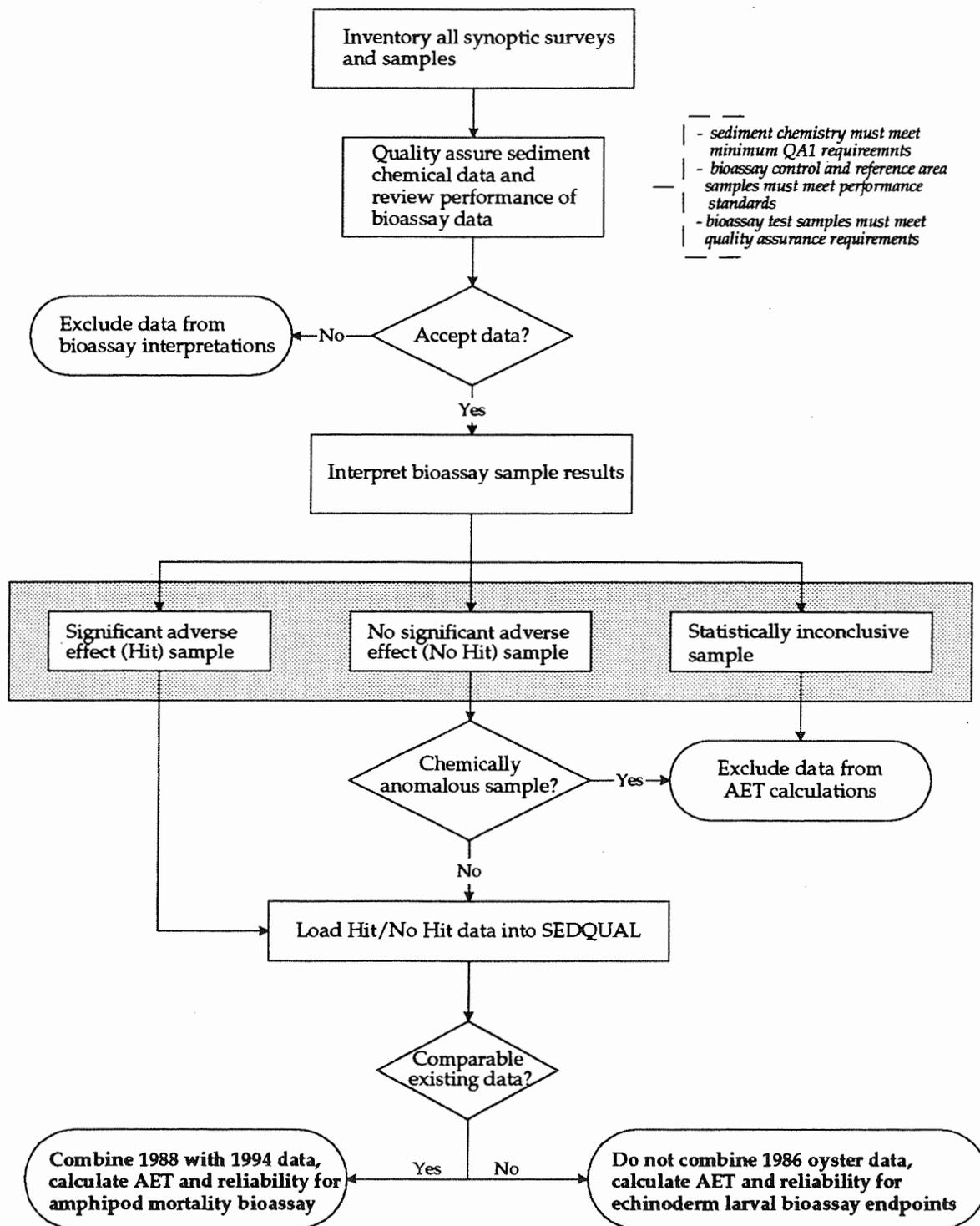
The data identification and acquisition, QA review and guidelines, bioassay performance standards, determination of biological effects, including the identification of inconclusive and anomalous samples, calculation of AETs and predictive reliability are described below.

### **Data Acquisition**

The 1994 re-evaluation of AETs depended in part on the acquisition of high quality synoptic data not used to calculate the 1988 AETs. In order to identify and obtain such data, PSDDA program staff contacted many groups and individuals during 1992 and 1993.

PSDDA agencies asked several federal agencies to identify and submit sediment quality data, including the U.S. Army Corps of Engineers - Seattle District (USACE), U.S. Environmental Protection Agency - Region 10 (EPA), U.S. Navy, and National Oceanic and Atmospheric Administration (NOAA)/National Marine Fisheries Service (NMFS). State and regional officials from various Ecology programs, the Departments of Health and Natural Resources, the Puget Sound Water Quality Authority (PSWQA), Seattle METRO and the University of Washington were also contacted. In addition, PSDDA agencies communicated

**Figure 2. General approach used to calculate 1994 Puget Sound AETs.** Quality assurance review ensures use of comparable data sets and excludes some samples from AET calculations. Bioassay interpretation yields "Hit", "No Hit" and "Inconclusive" samples (highlighted). Statistically inconclusive and anomalous samples are also excluded. The remaining biological effects data are used to calculate 1994 AET values.



with Jefferson, King, Pierce, Snohomish, Thurston, and Whatcom county staff, as well as port authorities, consulting firms and environmental laboratories.

The above groups were sent a standard letter or contacted by telephone. They were asked to identify all ambient sediment monitorings, dredging projects, compliance inspections and remedial or research investigations dating back to 1985 which involved collection of synoptic data. The responses to these inquiries were compiled into an inventory of synoptic surveys (Appendix B, Table B-1).

#### **Chemistry: Analytical Methods and Data Quality Assurance**

All chemical sediment quality data added to SEDQUAL and used in this AET re-evaluation were based on methods contained or identified in the Puget Sound Estuary Program (PSEP) Protocols and Guidelines (6).

Quality assurance guidelines for sediment chemistry data depend on the ultimate use of the data. Data that PSDDA agencies use for regulatory decisions on disposal of dredged material must meet or exceed "QA1" guidelines (6,16). Those guidelines verify lab and protocol performance by comparing analytical results from quality assurance samples to acceptable limits. Examples of QA samples include blanks, standard reference materials (SRM), duplicates, and matrix or surrogate spikes. Data used to develop or revise regulatory guidelines or standards must meet additional "QA2" data validation guidelines (17). QA2 guidelines include checking that the continuous calibration of analytical instruments and ensuring that calculations of final results are correct.

Chemical sediment quality data used to calculate the 1988 Puget Sound AETs met the more rigorous QA2 data validation guidelines. In contrast, the 1994 AET values presented in this report were calculated using survey data reflecting different levels of QA review. While all of those data met QA1 guidelines, only a subset was known to also meet QA2 guidelines. Many survey data did not undergo full QA2 validation because of unavailable or incomplete documentation.

To reconcile this inconsistency, those surveys containing samples setting new 1994 AETs, at a minimum, will undergo an *a posteriori* QA2 review<sup>10</sup>.

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<sup>10</sup> The PSDDA agencies agreed that recommendations to revise specific PSDDA ML or SL guidelines must consider results of that review.

## Bioassays: Analytical Methods and Data Quality Assurance

The 1994 AET re-evaluation focused exclusively on 10-day amphipod mortality and sediment larval effects as biological indicators of sediment toxicity. All bioassays which measured those effects were conducted following the basic PSEP Protocols (6). However, PSDDA survey samples may reflect slightly different bioassay protocols. That is because the PSDDA program has adopted minor modifications to the PSEP Protocols, based on the outcomes of two bioassay workshops (18,19) and its annual review process (8,9,11,12).

One protocol modification extended the acceptable bioassay sample holding time from two weeks to eight weeks (9). Thus, sediment bioassays samples from some PSDDA surveys were held longer before test initiation than 1988 AET samples. Other minor protocol modifications intended to improve test performance and interpretability included:

- measuring dissolved total ammonia and sulfides at the initiation and completion of each bioassay
- monitoring dissolved oxygen (DO) and pH daily
- aerating the overlying water if DO fell below 60% of saturation, or if high ammonia or total sulfides were present
- counting a minimum of 100 living larvae in control samples and a minimum of 20 abnormal larvae in reference and test sample replicates

The applicable sections of the PSEP Protocols (1989) are reproduced in Appendix A as Exhibit A-1. The protocol modifications made by the PSDDA agencies are summarized in Exhibit A-2.

PSDDA agencies carefully reviewed the bioassay data obtained since 1988 to ensure they were truly synoptic. Bioassay results linked to chemistry samples collected six months earlier or from a slightly different location, for example, were not used to calculate AETs. In such cases, the spatial and temporal variability observed in some Puget Sound sediments could result in high contaminant levels associated with no significant adverse biological effects, thereby setting anomalous AET values.

PSDDA agencies used both surface (0-2 cm) and subsurface (>2 cm) sediment samples for 1994 AET calculations, as long as they were synoptic and met all QA requirements. 1988 AETs were calculated using only synoptic surface sediment sample data.

Amphipod bioassays used the locally-collected marine species *Rhepoxinius abronius*. Calculations of sediment larval AETs were based on bioassays using

either bivalve (*Crassostrea gigas* or *Mytilus edulis*) or echinoderm larvae (*Dendraster excentricus* or *Strongylocentrotus droebachiensis* or *S. purpuratus*).

All new bioassay data underwent a careful, comprehensive review. PSDDA agencies examined the sample collection methods and holding times, test species, presence and performance of required control and reference samples, number of lab replicates, test duration and water quality for each bioassay batch. That information was compared to PSDDA-modified PSEP Protocols and requirements. Bioassay batches or samples were excluded from AET calculations if holding times were exceeded, or if there was no adequate negative control or reference sample.

### **Bioassays: Performance of Control and Reference Samples**

Positive control or reference toxicant bioassay data were available for nearly all new synoptic surveys. Those data were reviewed for a dose-response relationship and an  $EC_{50}/LC_{50}$  comparable to literature values. A few bioassay batches lacking a dose-responsive positive control were used in 1994 calculations, as long as other QA or performance standards were met. None of the batches in question contained only "Hit" samples or only "No Hit" samples, indicative of unusually sensitive or insensitive test organisms. Recent investigations support use of samples which lack only a dose-responsive positive control (20).

PSEP Protocols and the PSDDA program required a negative control sample to be run as part of each bioassay batch. All negative control sample data were compared to the existing performance standards presented in Table 1.

At least one reference sample per bioassay batch was also required. Reference samples had to be collected from a) recognized reference areas, such as Carr Inlet<sup>11</sup>, b) other areas meeting the description in the PSEP Protocols (6, page 18), or c) areas identified by recent reference area studies (21,22).

The existing performance standards for bioassay reference samples were based on the variability observed among the 1988 amphipod reference samples and PSDDA guidelines. Before comparing new reference sample data to those performance standards, PSDDA agencies incorporated the variability observed in the reference samples from the synoptic surveys collected since 1988. The result was a new performance standard for amphipod reference samples: a standard deviation of 18% among replicate mortalities (Appendix A, Figure A-1). The comparable reference standards for sediment larval abnormality and effective

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<sup>11</sup> Other established reference areas include Jetty Island, Samish and Sequim Bays.

mortality were standard deviations of 6% and 22%, respectively, (Appendix A, Figures A-2 and A-3).

Bioassay batches were excluded from 1994 AET calculations if their negative control or reference samples did not meet the performance standards in Table 1.

**Table 1.** Performance standards for bioassay control and reference samples used in 1994 re-evaluation of Puget Sound AETs.

Bioassay, Endpoint, (Test Duration)	Negative Control, Mean Value	Negative Control, Individ. Rep.	Reference Sample, Mean Value	Reference Sample, Std. Dev.
Amphipod mortality (10-day)	≤ 10%	≤ 20%	≤ 20%	≤ 18% <sup>a</sup>
Sediment larval abnormality (48 to 96-hour)	mortality ≤ 30% abnormality ≤ 10%	none	none	≤ 6% <sup>b</sup>
Sediment larval effective mortality (48 to 96-hour)	mortality ≤ 30%	none	≤ 35% <sup>c</sup>	≤ 22% <sup>d</sup>

- a. From the 95th percentile of standard deviations for mortality among 80 amphipod reference samples (Appendix A, Fig. A-1). The standard was 20% in 1988.
- b. From the 95th percentile of standard deviations for abnormality among 62 sediment larval reference samples (Appendix A, Fig. A-2). There was no comparable standard used for 1986 oyster abnormality AETs.
- c. Based on recommendations made by PSDDA in 1994 (12).
- d. From the 95th percentile of standard deviations for effective mortality among 62 sediment larval reference samples (Appendix A, Fig. A-3).

### Determination of Adverse Biological Effects

Prior to the 1988 update of Puget Sound AETs, technical experts discussed alternative methods of determining significant adverse effects, including the

appropriate statistical tests to use. This 1994 re-evaluation of AETs did not deviate substantially from the methods used in 1988 (4, pages 4-24 and Appendix C), believing that approach would preclude repeating earlier discussions and serve to maintain method consistency. The previous methods of determining significant adverse effects, and deviations from them, are highlighted in this section.

### Significant Amphipod Mortality

Determining significant adverse effects for all bioassays entailed comparing a mean response among lab replicates of individual test samples to the mean response among lab replicates of one or more reference area samples<sup>12</sup>. Those comparisons were usually straightforward, because most surveys involved a single reference sample per batch. For some surveys or batches, however, there was more than one acceptable reference sample.

In cases of multiple reference samples, PSDDA agencies used two methods to compare a test sample to those references. When possible, the mean amphipod mortality among lab replicates of a single test sample was compared to the mean mortality among lab replicates of a single reference sample having a similar sediment grain size. If a grain size match was not possible, then the mean test sample mortality was compared to the mean mortality of all reference samples. A recent investigation supported both methods of comparison (23).

The determination of significant adverse effects in the 10-day amphipod bioassay was done in five phases. Those phases are depicted in Appendix A, Figure A-4, and are described below.

- Phase I      A comprehensive QA review of control and reference sample data, as discussed in the previous section
- Phase II     Calculation of the mean percent mortality among lab replicates for all samples meeting the QA requirements. Test samples with a mean mortality less than 25% considered to have no significant adverse effects, and termed "No Hit" samples.
- Phase III    Comparison of all test samples with a mean mortality of 25% or greater to the mean mortality of one or more reference samples. Two-tailed F-test performed to determine if the variance among

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<sup>12</sup> Early AET calculations used negative control samples as alternative reference samples when the latter were lacking or they did not meet QA guidelines or performance standards. It was found that doing so greatly reduced the reliability of the resulting AETs. Therefore, bioassay batches without a valid reference sample were excluded from 1994 AET calculations.

samples was homogeneous. When variances homogeneous, comparison using standard two-sample, one-tailed t-test. When variances heterogeneous (rare), comparison using Satterthwaite approximate t-test (24).

Samples not statistically different from reference ( $p < 0.05$ ) and with a standard deviation among replicates of less than 15% considered "No Hit" samples. Those statistically different from reference considered "Hit" samples.

Phase IV Estimation of statistical power for samples having a mean mortality greater than 25%, not significantly different from reference sample and with replicate variability more than 15% (4, pages C16-22). Note: The addition of new amphipod bioassay test data did not change that 15% variability trigger (Appendix A, Figure A-5).

Calculation of statistical power of bioassay comparisons using a commercially available computer software program (25). Samples entering this phase of determination process considered "No Hit" samples if the comparison had a power value of at least 0.6. Those having a power value less than 0.6 were labeled "inconclusive" and not used in AET calculations.

Phase V Classification of samples as "Hit", "No Hit", or "Inconclusive." Determination of which "No Hit" samples were chemically anomalous (see Methods section, page 20).

### **Significant Sediment Larval Effects**

Calculations of 1994 sediment larval AET values were based only on bioassay data from surveys and samples obtained since 1988 AETs were calculated. All bioassays were conducted following PSDDA-modified PSEP Protocols, and included at least five laboratory replicates. Abnormality, mortality and effective mortality test endpoint data were entered following the conventions summarized in Appendix A, Exhibit A-2. They were used to calculate a percent response and significant difference from one or more reference samples.

PSDDA agencies determined significant adverse effects in the sediment larval bioassay using two of these different test endpoints. The larval abnormality endpoint was consistent with 1988 oyster larval AETs. However, the effective mortality (abnormality plus mortality) endpoint was consistent with current regulatory program endpoints.

## **Sediment Larval Abnormality**

Significant larval abnormality was determined as shown in Appendix A, Figure A-6. That determination was analogous to the one made for amphipod mortality, with two exceptions. First, no absolute threshold abnormality was required to define a "Hit" sample. In other words, a "Hit" based on abnormality alone was any sample with significantly greater abnormality ( $p < 0.05$ ) than an acceptable reference sample. Second, PSDDA agencies estimated the statistical power of each comparison involving high test sample variability. A standard deviation among test sample replicates exceeding 5% triggered that analysis. The 5% value was based on the 80th percentile of standard deviations among 303 sediment larval test samples (see Figure A-7). The second exception represented a departure from methods used in 1988.

## **Sediment Larval Effective Mortality**

Adverse effects, as indicated by effective mortality in the larval bioassay were determined as illustrated in Figure A-8 (Appendix A). Three alternative threshold levels were used to define a "Hit" and subsequently calculate AETs:

- test sample effective mortality 30% greater than and significantly different from reference sample effective mortality ( $p < 0.05$ )
- test sample effective mortality 15% greater than and significantly different from reference sample effective mortality ( $p < 0.05$ )
- test sample effective mortality significantly greater than reference sample effective mortality ( $p < 0.05$ )

The first of these thresholds reflected the PSDDA program's "single hit" threshold and the SMS biological standards for CSL, MCUL and  $SIZ_{max}$ . The second reflected the PSDDA program's "two hit" threshold and the SQS. The last was analogous to the abnormality determination, which lacked an absolute threshold.

For determining significant effective mortality in the sediment larval bioassay, power analysis was triggered by a standard deviation among test sample replicates of 22% (see Appendix A, Figure A-9).

## **Chemically Anomalous Samples**

PSDDA agencies identified chemically anomalous amphipod and sediment larval samples according to the "fixed factor difference" method - the same method selected in 1988 from among several options (4, page C-14). A sample was anomalous if it met the following conditions:

- it was determined to be a "No Hit" sample
- it had the greatest concentration of a given COC among all "No Hit" samples
- its concentration exceeded by a factor of three or more the concentration of the next highest ranked "No Hit" sample

Samples could be anomalous either for dry weight- or TOC-normalized chemical concentrations, or both. All samples identified as chemically anomalous were excluded from the corresponding AET calculations.

### Calculating AETs and Predictive Reliability

1994 AET values were calculated using the same general methods as were used in 1988 (4, *Appendix A*). All synoptic sediment samples which did not exhibit significant adverse effect, "No Hit" samples, were ranked from highest to lowest concentration for a single chemical of concern. The "No Hit" sample with the greatest concentration generally established the AET. If that sample was found to be chemically anomalous, then the sample with the next highest concentration set the AET. That AET value was confirmed by one or more "Hit" samples with a higher concentration. However, if no "Hit" sample had a greater concentration, then the AET was qualified as a minimum value with a "G" or by a ">" symbol. The determination of AETs was made for all chemicals of concern. The result was a group of AETs for a single biological indicator, interpretive endpoint, unit of measure and measurement basis.

In 1994, this AET calculation process was repeated to generate several AET groups. The four principal AET groups were for dry weight- and TOC-normalized amphipod mortality and echinoderm larval abnormality, reported either in units of ppm or ppb. AET groups were also combined and evaluated as a "suite." The 1988 suite of Puget Sound AETs was extended to include 1994 echinoderm larval values.

As was done in 1988 (4), a limited list of chemicals was removed from each group of AETs so that they could not contribute in any way to the predictive reliability of the 1994 AETs. The chemicals excluded from AET groups are listed in Appendix B (Table B-10). They primarily include compounds and elements which should not be implicated in biological effects (e.g., calcium or sodium) or conventional parameters which, though they may contribute to toxicity, might not represent broadly toxic anthropogenic pollutants (e.g., ammonia).

The reliability of 1994 AET groups was assessed using the same three measures as were used in 1988: sensitivity, efficiency and overall reliability (Figure 3). Sensitivity was defined as the percentage of correctly predicted "Hit" samples -- those samples which exceed at least one AET value and exhibit

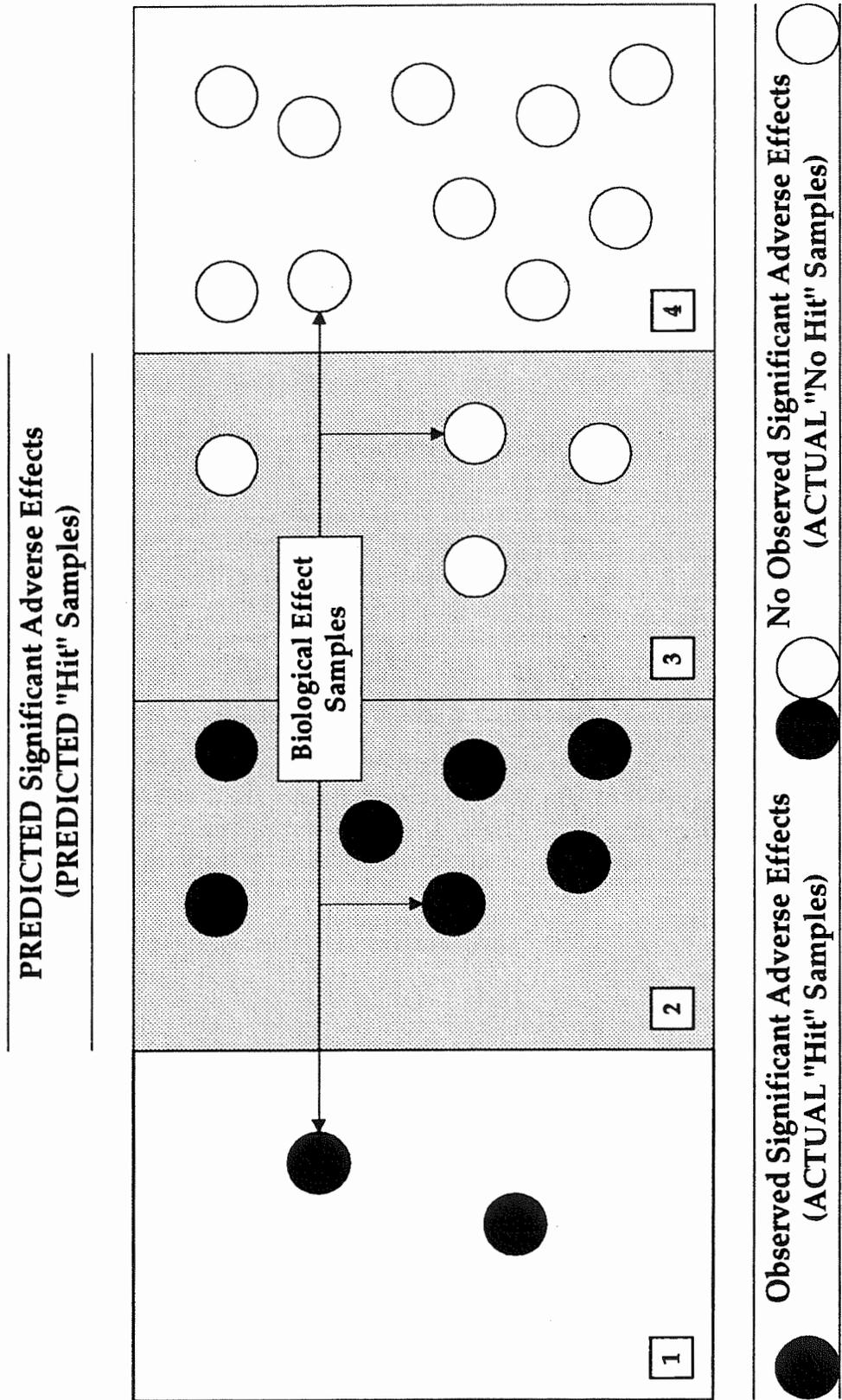
significant adverse effects. Efficiency was calculated as the percentage of all predicted "Hit" samples which actually exhibited significant adverse effects. In other words, efficiency was a measure of predictive accuracy. A highly efficient group of AETs predicted few "Hit" samples incorrectly. The last measure of reliability for a group of AETs, overall reliability, was computed as the percentage of all "Hit" and "No Hit" samples which were correctly predicted.

When the same data set used to calculate a group of AETs was compared to those AET values, efficiency was computed to be 100% by virtue of the AET definition. Any sample which exceeded the "No Hit" sample having the highest concentration for a given COC, thus setting that AET, was correctly predicted to be a "Hit" sample. Because efficiency calculated in that manner was not particularly useful, an "Independent" reliability analysis was sometimes conducted. That independent reliability analysis included a number of steps:

- temporarily withdrawing a single sample from a data set,
- computing AET values from the samples remaining,
- comparing the single sample to those AETs,
- recording whether the comparison predicted the sample which was temporarily withdrawn to be a "Hit" or a "No Hit" sample
- recording whether that prediction was correct
- repeating the process for all remaining samples (withdrawing a single sample, calculating AETs, comparing the single sample to those AETs, and so on)

Independent reliability calculations were conducted to allow meaningful comparisons of efficiency values for 1994 AET values to ones reported for corresponding 1988 AETs. Independent reliability analysis also enabled the predictive efficiency of various 1994 AET groups to be compared.

**Figure 3. Predictive reliability of Apparent Effects Threshold values.** Sensitivity is a measure of correctly predicted "Hit" samples. In this example, sensitivity = [(samples in box 2)/(samples in boxes 1+2)] X100 = 77%. Efficiency is a measure of accuracy of the "Hit" sample predictions [(box 2)/(boxes 2+3)] X100 = 63%. The overall reliability is the percentage of all predictions which is correct [(samples in boxes 2+4)/(all samples)] X100 = 74%.



The PSDDA agencies also calculated reliability after pooling all determinations of significant adverse effects. Any sample exhibiting at least one type of significant adverse effect was considered a "Hit" sample, even if no other adverse effects were found. The pooled reliability results showed how well a given group of AETs predicted samples exhibiting any type of significant adverse effect.

For this project, these three measures of reliability were also calculated across AET groups. For example, the sensitivity of the "LAET group" was calculated using only the lowest of the five AET values for each COC. The efficiency of the "HAET group" was similarly calculated using only the highest of five AET values.

## RESULTS

This section compares the final 1988 and 1994 Puget Sound AET databases, the former being a subset of the latter. It also describes why certain synoptic survey and sample data were excluded from AET calculations. It then focuses on the results of amphipod mortality and echinoderm larval abnormality AET calculations, as well as the predictive reliability of new AET values.

### Inventory of Synoptic Data for Puget Sound

The PSDDA agencies reviewed data from many dredging and non-dredging sediment surveys obtained since the "1988 Update and Evaluation of Puget Sound AET" (4). Most of the data met QA requirements and were entered into SEDQUAL. At the time this report was first drafted, there were 184 surveys in SEDQUAL which contained sediment chemistry data. Some 2970 stations and 3644 samples were geographically or temporally unique. From those, PSDDA agencies compiled an inventory of synoptic surveys and samples distinct from those used to calculate 1988 AETs. That inventory is presented in Table B-1 of Appendix B.

A total of 76 surveys contained 835 samples on which amphipod bioassays were conducted. Nine surveys and 168 samples involved sediment testing using bivalve larval species. There were 35 additional surveys with 316 samples which used echinoderm larvae as bioassay test organisms<sup>13</sup>.

### Data Excluded from AET Calculations

PSDDA agencies obtained and carefully reviewed most of those surveys. A few were excluded because they were not truly synoptic or failed to meet QA1 requirements. Numerous surveys, batches and samples were also excluded from 1994 AET calculations for other reasons. They are listed in Tables B-2 through B-5 and summarized below.

Of the 824 samples in the inventory, a total of 452 amphipod bioassay samples were not used in the 1994 AET calculations. The lack of at least one matching reference sample per batch was the most common reason for excluding amphipod test samples. That was true for 31 surveys and 162 samples. Seventy-six samples from three surveys were excluded because they failed to meet QA1 requirements. PSDDA agencies also excluded bioassay batches having poor reference or negative control sample performance (81 and 65 samples, respectively).

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<sup>13</sup> The count of synoptic samples may include retests of some batches or samples.

The next step was to determine which among the 393 remaining samples did and which did not exhibit significant adverse effects. Three of the "No Hit" samples were found statistically inconclusive (Table B-6). The remaining 390 samples were combined with the 295 used in 1988. From that subtotal, five samples were excluded from the 1994 inventory of synoptic surveys and samples, as well as six samples from the 1988 sediment quality values database. Those 11 samples were found to be chemically anomalous (Tables B-2 and B-7). That left 674 samples on which the dry weight-normalized 1994 amphipod mortality calculations were based. Ten samples were found to be anomalous when chemical concentrations were TOC-normalized.

The PSDDA agencies inventory identified nine surveys and 168 samples on which bivalve larval bioassays were conducted. Seventy-four samples from two surveys failed to meet QA1 requirements. Four surveys contained 63 samples for which there was no negative control meeting performance standards. Only three surveys met QA guidelines and negative control performance standards: DUWO&M90, PIER53BL, and PSREF90. Those surveys contained only 31 samples, an inadequate number upon which to base AET values (*4, page 50*).

Echinoderm larval bioassays were conducted on 35 surveys and 316 samples. Negative control sample results from four surveys and 45 samples did not meet performance standards. Matching reference samples were missing for five surveys and 30 samples. Two surveys containing 15 samples failed to meet other QA requirements. Reference samples for an additional two surveys (two samples) did not meet performance standards.

Eleven of the remaining 224 samples were found to be statistically inconclusive (Table B-4 and B6). Eight samples were excluded for being chemically anomalous (Table B-7). That left 205 samples on which 1994 dry weight-normalized echinoderm larval AETs were determined. One sample was found to be anomalous when chemical concentrations were TOC-normalized.

Neither the 1994 sediment bivalve larval nor the echinoderm larval effects data could be combined with the 1986 oyster larval effects data. The latter reflected substantially different protocols, used only two lab replicates, and determined the significance of adverse effects using a experiment-wise statistical test. In addition, the 1986 oyster AETs were based on geographically-limited data: two Commencement Bay surveys (CBMSQS, CBBLAIR).

#### **The Puget Sound AET Database: 1988 and 1994**

Sixty-two surveys, 505 stations and 590 synoptic samples were added to the AET database. Synoptic data from those samples were combined with data used in 1988 to form the basis for the analyses and results presented in this report.

Table 2 compares the final number of synoptic surveys and samples used to calculate AETs in 1994 to the number used in 1988. Amphipod mortality samples now total 674, while results from 29 surveys and 205 samples comprise the basis for echinoderm larval AETs.

## **Amphipod Mortality AETs**

### **Dry Weight-Normalized Values and Samples Setting Them**

Amphipod AETs for 28 PSDDA and SMS chemicals of concern remained the same as 1988 values, despite inclusion of additional synoptic data in 1994 calculations. Many AET values, however, did change. Those changes are apparent in the comparison of 1994 and 1988 amphipod AETs (Table 3) or can be discerned from the complete results presented in Appendix C (Tables C-1 through C-6).

Of the amphipod AET values which changed, all increased in magnitude except for the AET for benzyl alcohol. Those for trace metals and individual HPAHs were most affected. AETs for arsenic, cadmium, lead, mercury and zinc increased by factors ranging from 1.1 to 4.8. Values for chromium and nickel also increased, but were only estimated values, not exceeded by a "Hit" sample having a higher concentration. Six individual HPAH AETs increased by an average factor of 2.2. Values for LPAH, butyl benzyl phthalate, three substituted phenols (2-methyl phenol, 2,4-dimethyl phenol and pentachlorophenol) and p,p'-DDD increased by more modest factors of 1.1 to 1.5. In contrast, the 1994 amphipod AET for phenanthrene (21,000 ppm) was nearly triple the 1988 value, and appeared to drive the new AET for LPAH (29,000 ppm).

AET values for ethylbenzene, total xylenes and p,p'-DDT, only estimated in 1988, were confirmed by at least one 1994 "Hit" sample. Nickel and bis[2-ethylhexyl]phthalate AETs increased, but remained estimated values. Although based on limited data, amphipod AETs for hexachloroethane, trichloroethene, five pesticides, tributyl tin and other possible COCs could be calculated in 1994. Values were not reported for those chemicals in 1988.

The amphipod AET for benzyl alcohol was the only one found to decrease. It fell from 870 ppb in 1988 to 73 ppb in 1994.

Stations and samples used to calculate 1988 amphipod AETs dominated the list of those which established 1994 values (Table 3). That was because many of the 1988 AETs did not change and some of the new values were set by 1988 AET samples. The latter occurred because additional synoptic data caused a few of the 1988 AET samples to no longer be chemically anomalous. The re-introduction of those samples into 1994 calculations made it possible for them to establish new

**Table 2.** The Puget Sound sediment quality values (SEDQUAL) database: a comparison of synoptic surveys, stations and samples used to calculate AET values in 1988 and 1994. The number of samples with significant adverse effects ("Hit" samples) is given in parentheses. Note: SEDQUAL also contains extensive nonsynoptic data.

Biological Effect(s)	1988 Surveys/ Stations/Samples ("Hit" Samples) <sup>a</sup>	Added Surveys/ Stations/Samples ("Hit" Samples) <sup>b</sup>	1994 Surveys/ Stations/Samples ("Hit" Samples)
Amphipod mortality	9/284/287 (106)	38/329/385 (75)	47/613/674 <sup>c</sup> (181)
Benthic abundance	6/190/201 (108)		6/190/201 (108)
Echinoderm larval abnormality		24/176/205 (79)	24/176/205 (79)
Microtox luminosity	1/50/50 (29)		1/50/50 (29)
Oyster larval abnormality	2/56/56 (17)		2/56/56 (17)

- a. Taken from "Update and Evaluation of Puget Sound AET" (4)
- b. Synoptic surveys, stations and samples added to SEDQUAL for the 1994 re-evaluation of AETs (this report).
- c. The total number of amphipod mortality samples listed here exceeds the sum because two of the 1988 samples could no longer be excluded as chemically anomalous.

AETs. For example, the EBCHEM sample SS-10 was no longer anomalous in 1994 and set four amphipod AETs.

Samples from seven newly-obtained synoptic surveys established 17 of the new 1994 amphipod AETs. Samples from the SITCUMRI survey accounted for nine of those. One sample (SITCUMRI, 300112NM5) set new AETs for three metals and three PAHs. No other newly-obtained sample set more than two.

### TOC-Normalized Values and Samples Setting Them

Complete results of 1994 TOC-normalized amphipod AET calculations are presented in Appendix C (Tables 7-12). TOC-normalized values were less affected by additional synoptic data than the 1994 dry weight-normalized amphipod AETs (Table 4). Thirty-five remained the same in 1994. AETs for individual LPAHs, chlorinated organics, phthalates, phenols, miscellaneous extractables and volatile organics were particularly unaffected.

Eighteen of the recalculated TOC-normalized amphipod AETs differed from 1988 values. Trace metal and HPAH AETs accounted for more than half of those new values. The metals tended to decrease, with one remaining an estimate (chromium). TOC-normalized AETs for mercury and silver increased by 40% and 60%, respectively.

AETs for the HPAHs benzo[g,h,i]perylene, chrysene, dibenzo(a,h)anthracene and indeno(1,2,3-c,d)pyrene increased by an average of 40%. The amphipod TOC-normalized AET for phenanthrene increased, however, the increase was not extreme (690 ppm to 840 ppm) and did not affect the AET for LPAH. AETs for butyl benzyl phthalate and p,p'-DDD rose slightly.

TOC-normalized amphipod AETs for bis[2-ethylhexyl] phthalate and phenol increased from 1988 to 1994, but the new values were only estimates. In contrast, new data confirmed the AET for p,p'-DDT (16 ppm TOC) which could only be estimated in 1988. The TOC-normalized amphipod AET for benzyl alcohol decreased, as did the dry weight-normalized value. In 1994, TOC-normalized amphipod AETs could also be calculated for COCs not currently part of the PSDDA and SMS programs.

There was no lower or upper limit placed on TOC content of the samples setting the new AETs. However, anomalous "No Hit" samples were excluded based on their TOC-normalized chemistry (see Methods).

New TOC-normalized amphipod AET values were most often set by samples collected since the 1988 AETs were calculated (Table 4). Twelve samples from nine surveys caused a dozen AETs to change. Two of those samples

(SITCUMRI samples 300112NM5 and 300097NSE1A) set six new AETs. That was in contrast to seven new AETs set by eight samples from four older surveys.

### Overview of Sediment Larval AETs

The PSDDA agencies calculated sediment larval abnormality AETs using both sediment bivalve and echinoderm larval bioassay data obtained since 1988. Preliminary results of those calculations were presented at the 1993 PSDDA ARM and elsewhere (11, 18).

In response to those presentations, technical experts recommended calculating separate bivalve and echinoderm larval abnormality AETs<sup>14</sup>. However, in doing so, the PSDDA agencies found that the 1994 bivalve larval abnormality AETs would be based on synoptic data which a) was inadequate to calculate stable AETs<sup>15</sup>, b) lacked reference samples, and c) were not geographically representative of Puget Sound. Thus, bivalve larval AET calculations were not pursued further and are not presented in this report.

In order to be consistent with the 1986 oyster AETs, echinoderm larval AETs were calculated based on the abnormality endpoint alone. The PSDDA agencies also calculated echinoderm larval AETs based on effective mortality endpoints more consistent with current regulatory definitions of significant adverse effects. Those endpoints are described in the Methods section of this report.

Early results showed echinoderm larval AETs based on the effective mortality endpoint were much less sensitive than ones based on abnormality alone (Figure C-1). In other words, AETs based on "Hit" samples having any level of significantly greater effective mortality than a reference sample correctly predicted fewer observed adverse effects than echinoderm larval abnormality AETs.

Therefore, the remainder of this section focuses exclusively on the calculation of 1994 echinoderm larval abnormality AETs. Those values are the most sensitive echinoderm larval AETs and are most consistent with the 1986 oyster abnormality AETs. Nevertheless, echinoderm larval effective mortality AET and reliability results are presented in Appendix C (Tables C-25 through C-43).

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<sup>14</sup> Bivalve and echinoderm larvae were expected to differ in sensitivities to chemical pollutants.

<sup>15</sup> More than 50 stations or samples were required for calculated AET values to be less variable and more efficient (4, page 50).

## Echinoderm Larval Abnormality AETs

### Dry Weight-Normalized Values and Samples Setting Them

The 1994 echinoderm larval abnormality AETs were generally lower than the corresponding 1986 oyster AETs, whether dry weight-normalized (Table 5) or TOC-normalized (Table 6).

Dry weight-normalized echinoderm AETs for four trace metals decreased to levels averaging 35% of 1986 oyster AET values. Oyster abnormality AETs for antimony and silver were not reported in 1986, but were calculated using 1994 data. The chromium AET chromium was estimated to be at least 96 ppm. Only the AET for copper remained equal to that of the existing oyster AET.

Echinoderm abnormality AETs for twelve individual PAHs decreased to concentrations averaging 40% of their 1986 oyster counterparts. The summed parameters LPAH and HPAH were 23% and 46% of the original oyster AETs. Echinoderm AETs for phthalates were all lower than the corresponding oyster values. Three of those were confirmed values, while three were only estimated. Most of the phenol, miscellaneous extractable and volatile compounds were also lower.

AETs for nine chemicals of concern (mercury, nickel, silver, benzo[g,h,i]perylene, dibenzo(a,h)anthracene, indeno(1,2,3-c,d)pyrene, 2,4-dimethylphenol, pentachlorophenol, p,p'-DDT) increased, averaging 2.2 times the 1986 oyster values. The pentachlorophenol AET could only be estimated.

Confirmed echinoderm AET values for just six of the PSDDA or SMS chemicals of concern were greater than 1986 oyster AETs: mercury, silver, benzo[g,h,i]perylene, dibenzo(a,h)anthracene, indeno(1,2,3-c,d)pyrene, and 2,4-dimethylphenol.

Ten of the surveys obtained since 1988 contained 26 samples which set the echinoderm AETs (Table 5). Seven samples from the SITCUMRI survey set 25 of those values. One sample from the SITCUMRI survey (300018NSB1) set 12 AETs: five for trace metals, three for LPAHs, and four for HPAHs. Sample MET570XXS004 from the METROEBP survey set 5 AETs, including values for three HPAHs. A single PIERD\_91 sample (S8) also established five AETs, mostly from the pesticide and PCBs analytical group.

### TOC-Normalized Values and Samples Setting Them

TOC-normalized echinoderm larval abnormality AETs are listed in Table 6. Values for all trace metals except silver were less than the corresponding 1986

oyster AETs. 1994 echinoderm values for both LPAHs and HPAHs, however, were usually greater than the oyster analogues. Eleven of the remaining AETs could not be confirmed by a "Hit" sample with a higher concentration.

Twenty-five samples from 14 surveys established the TOC-normalized echinoderm abnormality AETs (Table 6). Twenty-six of those values were set by samples from just three surveys: METROEBP, NAVYMANC and TERMNL91. One METROEBP sample (MET570XXS004) was responsible for nine new AETs, including values for six individual HPAHs. The sample MANCHECCC002 (NAVYMANC survey) set five AETs for trace metals. A single TERMNL91 sample (TERMNL91S003) set seven TOC-normalized echinoderm AETs, including six for LPAHs.

### **Predictive Reliability of 1994 AETs**

The three measures of reliability used to evaluate AETs - "sensitivity", "efficiency", and "overall reliability" - were described in Methods and conceptually summarized in Figure 4. Sensitivity measures the ability of chemical AETs to correctly predict samples exhibiting significant adverse biological effects ("Hit" samples). Efficiency is the fraction of all "Hit" sample predictions which are correct. AETs which are highly efficient accurately predict "Hit" samples while yielding few false positive predictions. Overall reliability is the fraction of all predictions, both "Hit" and "No Hit" samples, which are correct.

Some reliability results for the 1994 amphipod mortality and echinoderm larval abnormality AETs are summarized in Table 7. The reliability of 1988 amphipod and 1986 oyster AETs are included for comparison purposes. Those results are organized by the AET type and means of chemical normalization: dry weight or TOC. Table 7 also lists some reliability analyses which are suggested in Recommendations for Future Work or which may be recommended by technical experts.

### **Amphipod Mortality AETs**

The sensitivity of dry weight-normalized amphipod AETs declined from 58% in 1988 to 43% in 1994. Efficiency, as indicated by independent reliability calculations, declined from 67% in 1988 to 52% in 1994. However, the overall reliability of 1994 values was similar to that found in 1988, 84% and 85%, respectively. The independent reliability calculation confirmed the fact that overall reliability was unchanged. The 1994 TOC-normalized amphipod AETs were also less sensitive than their 1988 counterparts: 36% in 1994 compared to 45% in 1988. Overall predictive reliability fell slightly, from 80% in 1988 to 78% in 1994.

**Table 3. Comparison of 1994 to 1988 dry weight-normalized amphipod mortality AET values.** New values are printed in bold italics. A ">" symbol indicates minimum values, not be confirmed by a "Hit" sample with a greater concentration. The ratio 1994 AET:1988 AET is given for confirmed values which changed. Stations and samples setting new 1994 AETs are also listed.

Chemical Group/ Chemical of Concern	Amphipod Mortality		1994: 1988		Survey Code and Station_ID	Sample_ID
	1994 AET	1988 AET	1988	1994		
<b>Metals (mg/kg or ppm)</b>						
Antimony	200	200				
Arsenic	<b>450</b>	93	4.8		EBCHEM, DR-12	DR-12
Cadmium	14	6.7	2.1		SITCUMRI, NM-5	300112NM5
Chromium	<b>&gt;1100</b>	270			EBCHEM, SS-10	SS-10
Copper	1,300	1,300				
Lead	<b>1,200</b>	660	1.8		SITCUMRI, NM-5	300112NM5
Mercury	2.3	2.1	1.1		PSDDA1, EBZ01	EBZ01C
Nickel	<b>&gt;370</b>	>140			EBCHEM, SS-10	SS-10
Silver	6.1	6.1				
Zinc	<b>3,800</b>	960	4.0		SITCUMRI, NM-5	300112NM5
<b>Organic Compounds (ug/kg or ppb)</b>						
<b>Low molecular weight PAH</b>						
LPAH	<b>29,000</b>	24,000	1.2		SITCUMRI, NM-5	300112NM5
2-Methylnaphthalene	1,900	1,900				
Acenaphthene	2,000	2,000				
Acenaphthylene	1,300	1,300				
Anthracene	13,000	13,000				
Fluorene	3,600	3,600				
Naphthalene	2,400	2,400				
Phenanthrene	<b>21,000</b>	6,900	3.0		SITCUMRI, NM-5	300112NM5

**Table 3. Comparison of 1994 to 1988 dry weight-normalized amphipod mortality AET values.** New values are printed in bold italics. A ">" symbol indicates minimum values, not be confirmed by a "Hit" sample with a greater concentration. The ratio 1994 AET:1988 AET is given for confirmed values which changed. Stations and samples setting new 1994 AETs are also listed.

Chemical Group/ Chemical of Concern	Amphipod Mortality		1994: 1988		Survey Code and Station_ID	Sample_ID
	1994 AET	1988 AET	1994	1988		
<b>High molecular weight PAH</b>						
HPAH	69,000	69,000				
Benz[a]anthracene	5,100	5,100				
Benzo[a]pyrene	<b>3,500</b>	3,000	1.2		EBCHEM, SS-10	SS-10
Benzo[ghi]perylene	<b>3,200</b>	1,400	2.3		EBCHEM, SS-04	SS-04
Benzofluoranthenes	<b>9,100</b>	7,800	1.2		EBCHEM, SS-10	SS-10
Chrysene	<b>21,000</b>	9,200	2.3		SITCUMRI, NM-5	300112NM5
Dibenzo(a,h)anthracene	<b>1,900</b>	540	3.5		EBCHEM, SS-04	SS-04
Fluoranthene	30,000	30,000				
Indeno(1,2,3-c,d)pyrene	<b>4,400</b>	1,800	2.4		EBCHEM, SS-04	SS-04
Pyrene	16,000	16,000				
<b>Chlorinated organic compounds</b>						
1,2,4-trichlorobenzene	51	51				
1,2-dichlorobenzene	>110	>110				
1,3-dichlorobenzene	>170	>170				
1,4-dichlorobenzene	120	120				
Hexachlorobenzene	130	130				
<b>Phthalates</b>						
Bis[2-ethylhexyl] phthalate	>8300	>3100			SED18903, 12	1
Butyl benzyl phthalate	970	900	1.1		EBCHEM, SS-04	SS-04
Di-n-butyl phthalate	1,400	1,400				
Di-n-octyl phthalate	>2100	>2100				

**Table 3. Comparison of 1994 to 1988 dry weight-normalized amphipod mortality AET values.** New values are printed in bold italics. A ">" symbol indicates minimum values, not be confirmed by a "Hit" sample with a greater concentration. The ratio 1994 AET:1988 AET is given for confirmed values which changed. Stations and samples setting new 1994 AETs are also listed.

Chemical Group/ Chemical of Concern	Amphipod Mortality		1994: 1988		Survey Code and Station ID	Sample ID
	1994 AET	1988 AET	1994	1988		
<b>Phthalates, cont'd</b>						
Diethylphthalate	>1200	>1200				
Dimethylphthalate	>1400	>1400				
<b>Phenols</b>						
2-methyl phenol	77	63	1.2		SITCUMRI, NC-S4	300048NCS4
2,4-dimethyl phenol	77	72	1.1		SITCUMRI, NC-S4	300048NCS4
4-methylphenol	3,600	3,600				
Pentachlorophenol	<b>400</b>	360	1.1		DUWO&M90, DUJ9003XX	DUWO&M90S003
Phenol	1,200	1,200				
<b>Miscellaneous Extractables</b>						
Benzyl alcohol	73	870	0.1		CBMSQS, HY-50	HY-50
Benzoic acid	760	760				
Dibenzofuran	1,700	1,700				
Hexachlorobutadiene	180	180				
Hexachloroethane	<b>140</b>	NA			CBMSQS, HY-24	HY-24
N-nitrosodiphenylamine	48	48				
<b>Volatile organics</b>						
Ethylbenzene	50	>50			CBMSQS, HY-17	H-17
Tetrachloroethene	>210	>210				
Xylene, Total	<b>160</b>	>160			CBMSQS, HY-17	H-17

**Table 3. Comparison of 1994 to 1988 dry weight-normalized amphipod mortality AET values. New values are printed in bold italics. A ">" symbol indicates minimum values, not be confirmed by a "Hit" sample with a greater concentration. The ratio 1994 AET:1988 AET is given for confirmed values which changed. Stations and samples setting new 1994 AETs are also listed.**

Chemical Group/ Chemical of Concern	Amphipod Mortality		1994: 1988		Survey Code and Station_ID	Sample_ID
	1994 AET	1988 AET	1994	1988		
<b>Pesticides and PCBs</b>						
Aldrin	9.5	NA			TERM5_91, TER50101	TER592C001
Chlordane	2.8	NA			TERM5_91, TER50101	TER592C001
Dieldrin	3.5	NA			BLGM_91A, BLGM09MC	BLGMBYXXC004
Heptachlor	1.5	NA			CBLAIR, B10	B10
p,p'-DDD	63	43	1.5		EBCHEM, SS-04	SS-04
p,p'-DDE	62	15	4.1		EBCHEM, DR-10	DR-10
p,p'-DDT	270	>270			EBCHEM, KG-06	EBCHEM KG-06
Total DDT	24	NA			PIERD_91, 11	C5
Total PCBs	3,100	3,100				
<b>Other</b>						
TBT	>180	NA			PSDDA1, EBB01	EBB01C
Trichloroethene	0.8	NA			EBCHEM, WW-18	WW-18

**Table 4. Comparison of 1994 to 1988 TOC-normalized amphipod mortality AET values for Puget Sound.**  
 New values are printed in bold italics. A ">" symbol indicates a minimum value, not confirmed by a "Hit" samp with a greater concentration. The ratio 1994:1988 AETs is given for confirmed values which changed. Stations and samples setting new 1994 AETs are also listed.

Chemical Group/ Chemical of Concern	Amphipod Mortality AET		1994:		Survey Code Station_ID	Sample_ID
	1994	1988	1988	1994		
<b>Metals (mg/kg organic carbon; ppm)</b>						
Antimony	15,000	>55,000	0.27		EBCHEM, WW-18 EBCHEM, WW-19 EBCHEM, WW-20	WW-18 WW-19 WW-20
Arsenic	32,000	32,000				
Cadmium	1,100	1,100				
Chromium	>130,000	>150,000	0.87		EBCHEM, MG-01	MG-01
Copper	100,000	100,000				
Lead	48,000	110,000			SITCUMRI, NM-5	300112NM5
Mercury	300	210	1.43		PIERD_91, 1	S1
Nickel	20,000	>41,000	0.49		NAVYMANC, MANC2006	MANCHEXXS007
Silver	270	170	1.59		SOPARK91, SPRK0302	SOPARK91C002
Zinc	150,000	210,000	0.71		SITCUMRI, NM-5	300112NM5

**Organic Compounds (mg/kg organic carbon; ppm)**

<b>Low molecular weight PAH</b>	
LPAH	2,200 2,200
2-Methylnaphthalene	>120 >120
Acenaphthene	200 200
Acenaphthylene	66 66
Anthracene	1,200 1,200
Fluorene	360 360
Naphthalene	220 220

**Table 4. Comparison of 1994 to 1988 TOC-normalized amphipod mortality AET values for Puget Sound.**  
 New values are printed in bold italics. A ">" symbol indicates a minimum value, not confirmed by a "Hit" sample with a greater concentration. The ratio 1994:1988 AETs is given for confirmed values which changed. Stations and samples setting new 1994 AETs are also listed.

Chemical Group/ Chemical of Concern	Amphipod Mortality AET		1994:1988		Survey Code		Sample_ID
	1994	1988	1994	1988	Station	ID	
<b>Low molecular weight PAH, cont'd</b>							
Phenanthrene	840	690	1.22		SITCUMRI, NM-5		300112NM5
<b>High molecular weight PAH</b>							
HPAH	5,300	5,300					
Benz[a]anthracene	270	270					
Benzof[a]pyrene	210	210					
Benzof[ghi]perylene	<b>100</b>	78	1.28		SITCUMRI, NS-E1A		300097NSE1A
Benzofluoranthenes	450	450					
Chrysene	840	460	1.83		SITCUMRI, NM-5		300112NM5
Dibenzo(a,h)anthracene	50	47	1.06		SITCUMRI, NS-E1A		300097NSE1A
Fluoranthene	3,000	3,000					
Indeno(1,2,3-c,d)pyrene	120	88	1.36		SED18903, 40		1
Pyrene	1,000	1,000					
<b>Chlorinated organic compounds</b>							
1,2,4-trichlorobenzene	1.8	1.8					
1,2-dichlorobenzene	>5.8	>5.8					
1,3-dichlorobenzene	>15	>15					
1,4-dichlorobenzene	9	9					
Hexachlorobenzene	4.5	4.5					
<b>Phthalates</b>							
Bis[2-ethylhexyl] phthalate	>550	78	7.05		SED18903, 12		1
Butyl benzyl phthalate	49	42	1.17		CGPIER35, PIER01MC		PIER35XXC001

**Table 4. Comparison of 1994 to 1988 TOC-normalized amphipod mortality AET values for Puget Sound.**  
 New values are printed in bold italics. A ">" symbol indicates a minimum value, not confirmed by a "Hit" samp with a greater concentration. The ratio 1994:1988 AETs is given for confirmed values which changed. Stations and samples setting new 1994 AETs are also listed.

Chemical Group/ Chemical of Concern	Amphipod Mortality AET		1994:	Survey Code	Sample_ID
	1994	1988	1988	Station_ID	
<b>Phthalates, cont'd</b>					
Di-n-butyl phthalate	260	260			
Di-n-octyl phthalate	58	58			
Diethylphthalate	>110	>110			
Dimethylphthalate	53	53			
<b>Phenols</b>					
2-methyl phenol	3.1	3.1			
2,4-dimethyl phenol	6.5	6.5			
4-methylphenol	780	780			
Pentachlorophenol	24	24			
Phenol	>440	440	1.10	EVCHEM, NG-14	NG-14G
<b>Miscellaneous Extractables</b>					
Benzyl alcohol	5	73	0.07	CBMSQS, BL-21	BL-21
Benzoic acid	>170	>170			
Dibenzofuran	>170	>170			
Hexachlorobutadiene	6.2	6.2			
Hexachloroethane	2.7	NA		CBMSQS, HY-24	HY-24
N-nitrosodiphenylamine	>11	>11			
<b>Volatile organics</b>					
Ethylbenzene	>3.8	>3.8			
Tetrachloroethene	>22	>22			
Xylene, Total	>12	>12			

**Table 4. Comparison of 1994 to 1988 TOC-normalized amphipod mortality AET values for Puget Sound.**  
 New values are printed in bold italics. A ">" symbol indicates a minimum value, not confirmed by a "Hit" samp with a greater concentration. The ratio 1994:1988 AETs is given for confirmed values which changed. Stations and samples setting new 1994 AETs are also listed.

Chemical Group/ Chemical of Concern	Amphipod Mortality AET		1994: 1988		Survey Code		Sample_ID
	1994	1988	1994	1988	Station_ID		
<b>Pesticides and PCBs</b>							
Aldrin	<i>0.56</i>	NA			TERM5_91, TER50101		TER592C001
Chlordane	<i>0.16</i>	NA			TERM5_91, TER50101		TER592C001
Dieldrin	<i>0.13</i>	NA			PSDDAM90, EB_Z01XX		EB90_Z01
Heptachlor	<i>&gt;0.11</i>	NA			CBBLAIR, B10		B10
p,p'-DDD	<i>3.1</i>	2.2	1.41		EBCHEM, DR-10		DR-10
p,p'-DDE	<i>6</i>	1	7.04		EBCHEM, DR-10		DR-10
p,p'-DDT	<i>16</i>	>16			EBCHEM, KG-06		KG-06
Total DDT	<i>1.4</i>	NA			PIERD_91, 11		C5
Total PCBs	190	190					
<b>Other</b>							
TBT	<i>&gt;18</i>	NA			PSDDA1, EBB01		EBB01C
Trichloroethene	<i>0.06</i>	NA			EBCHEM, WW-18		WW-18

**Table 5. Comparison of 1994 dry weight-normalized echinoderm abnormality AETs to corresponding 1986 oyster abnormality AET values. A ">" symbol indicates a minimum value, not confirmed by a "Hit" sample with a greater concentration. The ratio 1994 echinoderm AET:oyster 1986 AET is given for values which differ. Stations and samples setting new 1994 AETs are also listed.**

Chemical Group/ Chemical of Concern	Echinoderm Abnormality		Oyster Abnormality		AET Set By:		Sample_ID
	1994 AET	1986 AET	1994	1986	Survey Code and Station_ID	Station_ID	
<b>Metals (mg/kg or ppm)</b>							
Antimony	9.3	NA			SITCUMRI, NS-B6		300013NSB6
Arsenic	130	700	0.19		SITCUMRI, NS-B1		300018NSB1
Cadmium	2.7	9.6	0.28		SITCUMRI, NS-B1		300018NSB1
Chromium	>96	NA			OLYHARFC, OLYH26XX		OLYHFCXS023
Copper	390	390		0.65	SITCUMRI, NS-B1		300018NSB1
Lead	430	660	2.37		PIERD_91, 8		S8
Mercury	1.4	0.59	2.82		BLGM_91A, BLGM0606		BLGMBYXXC006
Nickel	110	39		>.56	SITCUMRI, NS-B1		300018NSB1
Silver	8.4	1,600		0.29	SITCUMRI, NS-B1		300018NSB1
Zinc	460						
<b>Organic Compounds (ug/kg or ppb)</b>							
<b>Low molecular weight PAH</b>							
LPAH	1,200	5,200	0.23		SITCUMRI, NS-B1		300018NSB1
2-Methylnaphthalene	64	670	0.10		SITCUMRI, NS-B6		300013NSB6
Acenaphthene	130	500	0.26		SITCUMRI, NS-B3		300017NSB3
Acenaphthylene	71	>560			METROEBP, METR0803		MET570XXS004
Anthracene	280	960	0.29		SITCUMRI, NS-B1		300018NSB1
Fluorene	120	540	0.22		SITCUMRI, NS-B3		300017NSB3
Naphthalene	230	2,100	0.11		OLYHARFC, OLYH25MC		OLYHFCXS012
Phenanthrene	660	1,500	0.44		SITCUMRI, NS-B1		300018NSB1

**Table 5. Comparison of 1994 dry weight-normalized echinoderm abnormality AETs to corresponding 1986 oyster abnormality AET values. A ">" symbol indicates a minimum value, not confirmed by a "Hit" sample with a greater concentration. The ratio 1994 echinoderm AET:oyster 1986 AET is given for values which differ. Stations and samples setting new 1994 AETs are also listed.**

Chemical Group/ Chemical of Concern	Echinoderm Abnormality		Oyster	AET Set By:		Sample_ID
	1994 AET	1986 AET	1986 AET	1994: 1986	Survey Code and Station_ID	
<b>High molecular weight PAH</b>						
HPAH	7,900	17,000	0.46		SITCUMRI, NS-B1	300018NSB1
Benz[a]anthracene	960	1,600	0.60		SITCUMRI, NS-B1	300018NSB1
Benzo[a]pyrene	1,100	1,600	0.69		SITCUMRI, NS-B1	300018NSB1
Benzo[ghi]perylene	920	720	1.28		METROEBP, METR0803	MET570XXS004
Benzofluoranthenes	1,800	3,600	0.50		SITCUMRI, NS-B1	300018NSB1
Chrysene	950	2,800	0.34		NAVYMANC, MANC1605	MANCHEXXS003
Dibenzo(a,h)anthracene	240	230	1.04		NAVYMANC, MANC1605	MANCHEXXS003
Fluoranthene	1,300	2,500	0.52		SITCUMRI, NS-A3	300031NSA3
Indeno(1,2,3-c,d)pyrene	760	690	1.10		METROEBP, METR0803	MET570XXS004
Pyrene	2,400	3,300	0.73		TERM5_91, TER50302	TER592CC002
<b>Chlorinated organic compounds</b>						
1,2,4-trichlorobenzene	>4.8	64			BLAIR_91, BL915921	BLAIR91XC021
1,2-dichlorobenzene	NA	50				
1,3-dichlorobenzene	>4.4	>170			TERM5_91, TER50101	TER592CC001
1,4-dichlorobenzene	NA	120				
Hexachlorobenzene	NA	230				
<b>Phthalates</b>						
Bis[2-ethylhexyl] phthalate	1,700	1,900	0.89		PIERD_91, 14	S14
Butyl benzyl phthalate	200	>470			OLYHARFC, OLYH07MC	OLYHFCXXS013

**Table 5. Comparison of 1994 dry weight-normalized echinoderm abnormality AETs to corresponding 1986 oyster abnormality AET values. A ">" symbol indicates a minimum value, not confirmed by a "Hit" sample with a greater concentration. The ratio 1994 echinoderm AET:oyster 1986 AET is given for values which differ. Stations and samples setting new 1994 AETs are also listed.**

Chemical Group/ Chemical of Concern	Echinoderm Abnormality		Oyster	AET Set By:		Sample_ID
	1994 AET	1986 AET	1986 AET	1994: 1986	Survey Code and Station_ID	
<b>Phthalates, cont'd</b>						
Di-n-butyl phthalate	>31	1,400			NAVYHPFC, NAVY20MC	NAVY89XXC007
Di-n-octyl phthalate	>98	>420			OLYHARFC, OLYH07MC	OLYHFCXXS013
					OLYHARFC, OLYH26XX	OLYHFCXXS023
Diethylphthalate	>62	>73			NAVYHPFC, NAVY20MC	NAVY89XXC021
Dimethylphthalate	85	160		0.53	SITCUMRI, NS-A2	300047NSA2
<b>Phenols</b>						
2-methyl phenol	55	63		0.87	SITCUMRI, NC-I12	300078NCI12
2,4-dimethyl phenol	55	29		1.90	SITCUMRI, NC-I12	300078NCI12
4-methylphenol	110	670		0.16	OLYHARFC, OLYH25MC	OLYHFCXXS012
Pentachlorophenol	>150	>140			PIERD_91, 14	S14
Phenol	>220	420			NAVYMANC, MANC04MC	MANCHEXXC003
<b>Miscellaneous Extractables</b>						
Benzyl alcohol	>12	73			PIERD_91, 2	S2
Benzoic acid	>31	650			DAYISL91, DAYI0101	DAYISL91C001
Dibenzofuran	110	540		0.20	OLYHARFC, OLHY07MC	OLYHFCXXS013
Hexachlorobutadiene	1.3	270		0.005	SITCUMRI, NS-B3	300017NSB3
Hexachloroethane	NA	NA			SITCUMRI, NC-I8	300084NCI8
N-nitrosodiphenylamine	>25	130			METROEBP, METR0803	MET570XXS004

**Table 5. Comparison of 1994 dry weight-normalized echinoderm abnormality AETs to corresponding 1986 oyster abnormality AET values. A ">" symbol indicates a minimum value, not confirmed by a "Hit" sample with a greater concentration. The ratio 1994 echinoderm AET:oyster 1986 AET is given for values which differ. Stations and samples setting new 1994 AETs are also listed.**

Chemical Group/ Chemical of Concern	Echinoderm		Oyster		AET Set By:		Sample_ID
	Abnormality 1994 AET	Abnormality 1986 AET	Abnormality 1994	Abnormality 1986	Survey Code and Station_ID	Station_ID	
<b>Volatile organics</b>							
Ethylbenzene	4.0	37	0.11		SITCUMRI, NS-B6		300013NSB6
Tetrachloroethene	>1	140			SITCUMRI, NC-I12		300078NCI12
					SITCUMRI, NC-I8		300084NCI8
Xylene, Total	21	120	0.18		SITCUMRI, NS-B6		300013NSB6
<b>Pesticides and PCBs</b>							
Aldrin	10	NA			TERM5_91, TER50101		TER592C001
Chlordane	>4.5	NA			PIERD_91, 2		S2
Dieldrin	1.9	NA			PIERD_91, 6		C3
Heptachlor	2	NA			BLGM_91A, BLGM6518		BLGMBYXXC018
p,p'-DDD	28	NA			PIERD_91, 8		S8
p,p'-DDE	9.3	NA			PIERD_91, 8		S8
p,p'-DDT	12	>6	2.00		PIERD_91, 11		C5
Total DDT	37	NA			PIERD_91, 8		S8
Total PCBs	450	1,100	0.41		PIERD_91, 8		S8

**Table 6. Comparison of 1994 TOC-normalized echinoderm abnormality AETs to corresponding 1986 oyster abnormality AET values. A ">" symbol indicates a minimum value, not confirmed by a "Hit" sample with a greater concentration. The ratio 1994 echinoderm AET:oyster 1986 AET is given for values which differ. Stations and samples setting new 1994 AETs are also listed.**

Chemical Group/ Chemical of Concern	Echinoderm Abnormality		Oyster	AET Set By:		Sample_ID
	1994 AET	1986 AET	1986 AET	1994: 1986	Survey Code and Station_ID	
<b>Metals (mg/kg organic carbon; ppm)</b>						
Antimony	>2,100	3,300			NAVYMANC, MANC0702	MANCHEXXC002
Arsenic	5,800	88,000	0.07		NAVYMANC, MANC0702	MANCHEXXC002
Cadmium	>430	1,200			NAVYMANC, MANC2207	MANCHEXXC007
Chromium	>5,400	NA			OLYHARFC, OLYH02MC	OLYHFCXXC006
Copper	30,000	49,000	0.61		NAVYMANC, MANC0702	MANCHEXXC002
Lead	22,000	66,000	0.33		BLAIR_91, BL9140MC	BLAIR91XC026
Mercury	71	210	0.34		PIERD_91, 11	C5
Nickel	>49000	NA			NAVYMANC, MANC0702	MANCHEXXC002
Silver	270	>100			SOPARK91, SPRK0302	SOPARK91C002
Zinc	60,000	>200,000			NAVYMANC, MANC0702	MANCHEXXC002
<b>Organic Compounds (mg/kg organic carbon; ppm)</b>						
<b>Low molecular weight PAH</b>						
LPAH	22	370	0.06		PIERD_91, 11	C5
2-Methylnaphthalene	>53	NA			TERMNL91, T91_03XX	TERMNL91S003
Acenaphthene	>110	16			TERMNL91, T91_03XX	TERMNL91S003
Acenaphthylene	>18	>27				
Anthracene	93	>79			TERMNL91, T91_03XX	TERMNL91S003
Fluorene	73	23	3.17		TERMNL91, T91_03XX	TERMNL91S003
Naphthalene	>190	99			TERMNL91, T91_03XX	TERMNL91S003
Phenanthrene	140	120	1.17		METROEBP, METR0803	MET570XXS004

**Table 6. Comparison of 1994 TOC-normalized echinoderm abnormality AETs to corresponding 1986 oyster abnormality AET values. A ">" symbol indicates a minimum value, not confirmed by a "Hit" sample with a greater concentration. The ratio 1994 echinoderm AET:oyster 1986 AET is given for values which differ. Stations and samples setting new 1994 AETs are also listed.**

Chemical Group/ Chemical of Concern	Echinoderm Abnormality		Oyster	AET Set By:		Sample ID
	1994 AET	1986 AET	1986 AET	1994: 1986	Survey Code and Station ID	
<b>High molecular weight PAH</b>						
HPAH	150	960		0.16	PIERD_91, 8	S8
Benz[a]anthracene	170	110		1.55	METROEBP, METR0803	MET570XXS004
Benzo[a]pyrene	230	99		2.32	METROEBP, METR0803	MET570XXS004
Benzo[ghi]perylene	>240	31		1.35	METROEBP, METR0803	MET570XXS004
Benzofluoranthenes	310	230		1.35	TERMINL91, T91_03XX	TERMINL91S003
Chrysene	220	110		2.00	METROEBP, METR0803	MET570XXS004
Dibenzo(a,h)anthracene	48	120		0.40	NAVYMANC, MANC1705	MANCHEXXS004
Fluoranthene	320	160		2.00	NAVYMANC, MANC18XX	MANCHEXXS005
Indeno(1,2,3-c,d)pyrene	>190	33			METROEBP, METR0803	MET570XXS004
Pyrene	520	>210			METROEBP, METR0803	MET570XXS004
<b>Chlorinated organic compounds</b>						
1,2,4-trichlorobenzene	>2.4	2.7			BLAIR_91, BL915921	BLAIR91XC021
1,2-dichlorobenzene	NA	2.3				
1,3-dichlorobenzene	>0.26	>15			TERM5_91, TER50101	TER592C001
1,4-dichlorobenzene	NA	3.1				
Hexachlorobenzene	NA	9.6				
<b>Phthalates</b>						
Bis[2-ethylhexyl] phthalate	130	60		2.17	NAVYMANC, MANC0101	MANCHEXXC001
Butyl benzyl phthalate	5.2	>9.2			OLYHARFC, OLYH07MC	OLYHFCXXS013
Di-n-butyl phthalate	0.88	260		0.00	PSDDAM92, PMONS02	PMONS02S002

**Table 6. Comparison of 1994 TOC-normalized echinoderm abnormality AETs to corresponding 1986 oyster abnormality AET values. A ">" symbol indicates a minimum value, not confirmed by a "Hit" sample with a greater concentration. The ratio 1994 echinoderm AET:oyster 1986 AET is given for values which differ. Stations and samples setting new 1994 AETs are also listed.**

Chemical Group/ Chemical of Concern	Echinoderm		Oyster		AET Set By:	
	Abnormality 1994 AET	Abnormality 1986 AET	1994: 1986	Survey Code and Station_ID	Sample_ID	
<b>Phthalates, cont'd</b>						
Di-n-octyl phthalate	>0.55	>57		AMPRES92, APL_01MC	AMPRES92C002	
Diethylphthalate	>0.27	>5.3		NAVYHPFC, NAVY20MC	NAVY89XXC025	
Dimethylphthalate	NA	>22				
<b>Phenols</b>						
2-methyl phenol	>2.1	3.1		OLYHARFC, OLYH17MC	OLYHFCXXC001	
2,4-dimethyl phenol	NA	>1.3				
4-methylphenol	4.7	37	0.13	OLYHARFC, OLYH17MC	OLYHFCXXC001	
Pentachlorophenol	>9.3	>11		PIERD_91, 6	C3	
Phenol	3.2	>39		NAVYMANC, MANC18XX	MANCHEXXS005	
<b>Miscellaneous Extractables</b>						
Benzyl alcohol	>0.71	5		PIERD_91, 2	S2	
Benzoic acid	>2.0	>170		DAYISL91, DAY10101	DAYISL91C001	
Dibenzofuran	57	15	3.80	TERMINL91, T91_03XX	TERMINL91S003	
Hexachlorobutadiene	NA	11				
Hexachloroethane	NA	NA				
N-nitrosodiphenylamine	>6.4	>11		METROEBP, METR0803	MET570XXS004	
<b>Volatile organics</b>						
Ethylbenzene	NA	>3.8				
Tetrachloroethene	NA	>22				
Xylene, Total	0.15	>12		NAVYHPIL, EVNV03MC	NAVYII90C002	

**Table 6. Comparison of 1994 TOC-normalized echinoderm abnormality AETs to corresponding 1986 oyster abnormality AET values. A ">" symbol indicates a minimum value, not confirmed by a "Hit" sample with a greater concentration. The ratio 1994 echinoderm AET:oyster 1986 AET is given for values which differ. Stations and samples setting new 1994 AETs are also listed.**

Chemical Group/ Chemical of Concern	Echinoderm Oyster		AET Set By:		Sample_ID
	Abnormality 1994 AET	Abnormality 1986 AET	1994: 1986	Survey Code and Station_ID	
<b>Pesticides and PCBs</b>					
Aldrin	>0.56	NA		TERM5_91, TER50101	TER592C001
Chlordane	>0.26	NA		PIERD_91, 2	S2
Dieldrin	0.28	NA		PIERD_91, 6	C3
Heptachlor	>0.40	NA		BLGM_91A, BLGM3813	BLGMBYXXS002
p,p'-DDD	1.6	NA		PIERD_91, 16	C6
p,p'-DDE	>7.3	NA		PIERD_91, 16	C6
p,p'-DDT	>0.71	NA		PIERD_91, 11	C5
Total DDT	8.8	NA		PIERD_91, 16	C6
Total PCBs	18	>46		PIERD_91, 8	S8

## Echinoderm Larval Abnormality AETs

The sensitivity of 1994 dry weight-normalized echinoderm larval abnormality AET values was 48%, much lower than the 88% value recorded for oyster larval abnormality AETs in 1986 (Table 7). Using the independent reliability calculation, the PSDDA agencies found the efficiency (54%) and overall reliability (67%) of the echinoderm AETs to exceed the values for 1986 oyster AETs (37 % and 50%, respectively). Reliability results for TOC-normalized larval AETs were: 46% sensitivity and 79% overall reliability. Corresponding values for 1986 oyster AETs were 71% and 91%.

## Some Characteristics of Incorrectly Predicted Stations

The direct cause of low sensitivity was that "Hit" samples were not predicted to exhibit significant adverse effects because their chemistry did not exceed any AET value. One reason that may have occurred was that the effects observed in incorrectly predicted "Hit" samples were in response to sample handling or due to toxicity from conventional parameters, such as sulfides. A brief examination of the latter possibility is presented below.

The PSDDA agencies preliminarily examined concentrations of some sediment conventionals in "Hit" samples which were incorrectly predicted by 1994 AETs. A large fraction of those incorrectly predicted samples had "elevated" levels of bulk ammonia, percent fines, or total bulk sulfides<sup>16</sup> (Table 8). Percent clay and TOC were less frequently elevated. Nearly one-third of all incorrectly predicted dry weight-normalized amphipod "Hit" samples (31/104) showed elevated levels of at least two conventional parameters. One-half (24/47) of the incorrectly predicted echinoderm "Hits" fell into that category.

The PSDDA agencies considered excluding amphipod "Hit" samples having greater than 80% fines from 1994 AET calculations. They also considered another approach to reducing the number of samples in which fines content likely contributed to amphipod mortality. Modelled after DeWitt et al (26), that approach was to use the relationship observed in Puget Sound between sample percent fines and amphipod mortality to correct the mean sample mortality results. That correction, performed prior to determination of significant adverse effects, could potentially have reduced the frequency of incorrectly predicted "Hit" samples.

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<sup>16</sup> For this initial assessment of conventional parameters associated with incorrectly predicted "Hit" samples, "elevated" bulk ammonia concentrations were operationally defined to be > 20 ppm, "elevated" fines were > 80%, and "elevated" bulk sulfides were > 50 ppm. Fine grained sediment is defined by PSEP (6) to be the sum of percent silt (0.063 - 0.004 mm particles) and percent clay (less than 0.004 mm).

**Table 7. The predictive reliability of amphipod mortality and echinoderm larvae abnormality AETs.** 1994 AETs (shaded) are compared to original 1988 amphipod and 1986 oyster AETs. "Sensitivity," "Efficiency," "Overall Reliability," "Independent AETs," and "Pooled" are defined in the Background and Glossary sections of this report. Measures of reliability are given as percentages and fractions. Historical reliability values are from "1988 Update and Evaluation of Puget Sound AET" (4).

AET Group	Database for Comparison	Total Samples	Sensitivity	Efficiency	Overall Reliability
<b>Dry Weight Normalized</b>					
1986 Amphipod AETs	1986 AMPT	287	56% (59/106)	69% (59/86)	74% (213/287)
1988 Amphipod AETs	1988 AMPT	287	58% (62/106)	100% (62/62)	85% (243/287)
1988 Independent AETs	1988 AMPT	287	57% (60/106)	67% (60/90)	74% (211/287)
1988 Amphipod AETs	1994 AMPT	674	48% (87/181)	38% (87/227)	65% (438/674)
1994 Amphipod AETs	1994 AMPT	674	43% (79/181)	100% (79/79)	84% (569/674)
1994 Independent AETs	1994 AMPT	674	42% (76/181)	52% (76/146)	74% (499/674)
1988 Amphipod AETs	1994 Pooled	674	39% (119/309)	47% (119/251)	52% (352/674)
1994 Amphipod AETs	1994 Pooled	674	31% (95/309)	88% (95/108)	66% (447/674)
1986 Oyster AETs	1986 OYST	56	88% (15/17)	100% (15/15)	96% (54/56)
1986 Independent AETs	1986 OYST	56	88% (15/17)	37% (15/41)	50% (28/56)
1986 OYST AETs	1994 ECHN	205	75% (59/79)	60% (59/98)	71% (146/205)
1994 Echinoderm AETs	1994 ECHN	205	48% (38/79)	100% (38/38)	80% (164/205)
1994 Independent AETs	1994 ECHN	205	46% (36/79)	54% (36/67)	67% (137/205)
1986 OYST AETs	1994 Pooled	205	67% (206/309)	58% (206/358)	NA
1994 Echinoderm AETs	1994 Pooled	205	67% (206/309)	59% (206/348)	NA

**Table 7. The predictive reliability of amphipod mortality and echinoderm larvae abnormality AETs.** 1994 AETs (shaded) are compared to original 1988 amphipod and 1986 oyster AETs. "Sensitivity," "Efficiency," "Overall Reliability," "Independent AETs," and "Pooled" are defined in the Background and Glossary sections of this report. Measures of reliability are given as percentages and fractions. Historical reliability values are from "1988 Update and Evaluation of Puget Sound AET" (4).

AET Group	Database for Comparison	Total Samples	Sensitivity	Efficiency	Overall Reliability
<b>TOC Normalized</b>					
1988 Amphipod AETs	1988 AMPT	287	45% (48/106)	100% (48/48)	80% (229/287)
1988 Amphipod AETs	1994 AMPT	671	34% (61/181)	46% (61/133)	71% (476/671)
1994 Amphipod AETs	1994 AMPT	478	36% (58/162)	100% (58/58)	78% (368/478)
1994 Independent AETs	1994 AMPT	478	NA	NA	NA
1988 Amphipod AETs	1994 Pooled	671	28% (87/309)	52% (87/168)	55% (369/671)
1994 Amphipod AETs	1994 Pooled	478	NA	NA	NA
1986 Oyster AETs	1986 OYST	56	71% (12/17)	100% (12/12)	91% (51/56)
1986 OYST AETs	1994 ECHN	205	20% (16/79)	59% (16/27)	58% (119/205)
1994 Echinoderm AETs	1994 ECHN	205	46% (36/79)	100% (36/36)	79% (162/205)
1994 Independent AETs	1994 ECHN	205	NA	NA	NA
1986 OYST AETs	1994 Pooled	205	28% (88/309)	59% (88/148)	NA
1994 Echinoderm AETs	1994 Pooled	205	NA	NA	NA

**Table 7. The predictive reliability of amphipod mortality and echinoderm larvae abnormality AETs.** 1994 AETs (shaded) are compared to original 1988 amphipod and 1986 oyster AETs. "Sensitivity," "Efficiency," "Overall Reliability," "Independent AETs," and "Pooled" are defined in the Background and Glossary sections of this report. Measures of reliability are given as percentages and fractions. Historical reliability values are from "1988 Update and Evaluation of Puget Sound AET" (4).

AET Group	Database for Comparison	Total Samples	Sensitivity	Efficiency	Overall Reliability
<b>Mixed Normalization</b>					
1988 Amphipod AET		287	55% (58/106)	100% (58/58)	83% (239/287)
1994 Amphipod AET		478	NA	NA	NA
1994 Independent AET		478	NA	NA	NA
1994 Amphipod AET, Pooled		478	NA	NA	NA
1986 Oyster AET		56	88% (15/17)	100% (15/15)	96% (54/56)
1994 Echinoderm AET		205	NA	NA	NA
1994 Independent AET		205	NA	NA	NA
1994 Echinoderm AET, Pooled		205	NA	NA	NA