

PTI

ENVIRONMENTAL SERVICES

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DATA REPORT

**Puget Sound Estuarine Studies
Procedures for Monitoring Salmon Marine Net-Pens**

Submitted to

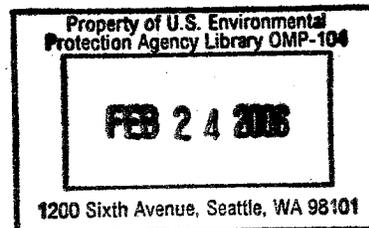
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LIST OF ACRONYMS

BOD	biological oxygen demand
CCB	continuing calibration blank
CCV	continuing calibration verification
COD	chemical oxygen demand
DQO	data quality objective
EDTA	ethylenediaminetetraacetic acid
EPA	U.S. Environmental Protection Agency
ICB	initial calibration blank
ICV	initial calibration verification
LCS	laboratory control sample
MRL	method reporting limit
NTU	nephelometric turbidity units
PHA	poly-hydroxy alkanate
PHB	poly- β -hydroxybutyrate
PLFA	phospholipid fatty acid
PSEP	Puget Sound Estuary Program
QA/QC	quality assurance/quality control
QAPP	quality assurance project plan
RPD	relative percent difference
RSD	relative standard deviation
SOW	statement of work
TKN	total Kjeldahl nitrogen
TOC	total organic carbon
TON	total organic nitrogen
TVS	total volatile solids

PTOC?

1.0 INTRODUCTION

The culturing of salmon in marine net-pens results in the release of solid wastes (fish feces and unconsumed food) to the surrounding aquatic environment. These wastes can result in increased concentrations of phytoplankton nutrients within the water column and increased bacterial activity among bottom sediments. In the case of bottom dwelling organisms, increased input of organic carbon and associated bacterial activity can produce anoxic conditions that dramatically affect the species composition and abundances of organisms.

The U.S. Environmental Protection Agency (EPA) Region 10 undertook a study of procedures for monitoring the near-field benthic effects of organic deposition from salmon marine net-pens. Sediments and infaunal macroinvertebrates were sampled at five net-pens in Puget Sound, Washington, during the spring of 1991.

Benthic samples were collected by SCUBA divers at each of five salmon net-pen facilities located at ScanAm #1 (Cypress Island, near Anacortes), Sea Farms WA #1 (Port Angeles), Global Aqua #2 (Rich Passage, near Bainbridge Island) and Global Aqua #3 (Clam Bay, near Manchester), and Paradise Bay Seafarms (Port Townsend). Samples were processed in the field and subsequently analyzed for sulfide, grain size, sediment chemistry, and benthic infauna.

One of the five net-pen operations (Paradise Bay Seafarms, Port Townsend) was also sampled using a surface vessel, the research vessel (R/V) *Kittiwake*. Six Port Townsend stations and a reference location were sampled by the R/V *Kittiwake*, with five replicate chemistry and benthic infauna replicate samples collected at each station. Sediments were also collected at each station for analysis of bacterial biomass (i.e., phospholipid). Water samples were collected at two stations.

This report contains the data resulting from these studies. The report is divided into six sections:

- Introduction
- Cruise report for surface vessel sampling
- Cruise report for diver sampling
- Sediment and water chemistry quality assurance/quality control (QA/QC) review
- Benthic infauna QA/QC review.

Eight appendices are also included. Appendix A includes sediment bacterial biomass data and data interpretation. Appendix B contains field notes from the diver sampling events. Appendix C contains all sediment and water chemistry data. Appendix D lists laboratory holding times for all chemistry analyses. Results of the benthic infauna analyses are included in Appendix E, and copies of all chain-of-custody forms are in Appendix F. Appendix G contains field notes from the vessel sampling event and Appendix H contains the EPA divers' field reports.

2.0 CRUISE REPORT PORT TOWNSEND VESSEL SAMPLING

The salmon net-pen monitoring survey was conducted during May 1-3, 1991, aboard the *R/V Kittiwake*. Samples were collected on a transect that extended from the north end of the Paradise Bay Seafarms fish pens in Port Townsend Harbor. Samples were also collected from a reference area near Port Townsend. Station locations are shown in Figure 1. Sediment chemistry, benthic infauna, and bacteria samples were collected at seven stations, and water quality was measured at two stations. The sediment chemistry portion of this study is described in detail in Chapter 4.0 of this report.

Two sizes of sampling gear were used to collect samples, a 0.025 m² Van Veen sampler and a dual 0.1 m² Van Veen grab sampler. Five replicate casts were taken at each station using both samplers (total 10 casts). Each of the five 0.025 m² Van Veen samples was sieved onboard the vessel on a 0.5 mm sieve using gentle streams of sea water. The resulting benthic infauna samples were transferred to sample containers and preserved with 10 percent buffered formalin.

Each cast with the larger ^{dual} Van Veen sampler yielded two 0.1 m² grabs. One of these grab samples was processed for benthic infauna while the second was used for chemical analyses. In the case of benthic infauna samples, sediments were sieved using two stacked sieves (1.0 mm and 0.5 mm) with the resulting infauna samples transferred to sample containers and preserved with 10 percent buffered formalin. The overlying water was siphoned off the sediment chemistry sample and sediments from the upper 2 cm were transferred directly to a sample container for sulfide analyses. The remaining upper 2 cm of sediment was subsequently placed in a stainless steel bowl, mixed thoroughly, and then transferred to sample containers for other laboratory analysis. Specific chemical analyses included BOD, COD, total phosphorous, total organic nitrogen, sulfides, total organic carbon, and total volatile solids. Analytical methods are described in Section 4.

Ten additional ^{location} marine sediment samples collected near the salmon net-pens during the Port Townsend vessel sampling event were sent to Oak Ridge National Laboratory. Field triplicate samples were submitted for the three stations (PTV1, PTV5, and PTV6) along with one sample from the reference area. The samples were analyzed for fatty-acids to quantitatively define the biomass, community structure, and nutritional status of the associated microbiota. The standard techniques used by the Institute for Applied Microbiology were followed (see Appendix A for detailed description). The specific analyses conducted were poly β-hydroxy alkanate (PHA) and gas chromatography/mass spectrometry for

phospholipid ester-linked fatty acids (PLFA). The methods, resulting data, and interpretation were submitted by the laboratory in report form. The report is included as Appendix A.

In general, the cruise was conducted efficiently and no problems were encountered. Excellent weather conditions prevailed throughout the cruise. The weather conditions facilitated accurate vessel repositioning once a station was established and minimized transit time between the fish pens and the reference area. A summary of the sampling activities that occurred during each day is presented in Table 1. A summary of the samples collected at each station is presented in Table 2. Station coordinates and distances from the commercial fish pens are provided in Table 3. All station and sample logs are on file at PTI.

Appendix
B
E
H

Sample identifiers used in the field were established as follows. The first three letters of the sample name represented the site name and survey type (i.e., PTV - Port Townsend vessel survey). The next character represented the station number (1 through 6). Following the station number was a letter designating the sample type (e.g., C - chemistry, B - benthic infauna, W - water, or P - bacterial phospholipid). Field replicate numbers followed the sample type. Sample identifier PTV3C2, for example, was the second chemistry field replicate collected at Station 3 during the vessel survey.

Benthic infauna samples required additional identifiers since two sizes of sampling gear were used at each station (i.e., 0.025- and 0.1-m² van Veen grab samplers) and all samples collected using the larger van Veen sampler were sieved on both 0.5-mm and 1.0-mm mesh sieves. Examples are as follows:

- PTV6BL15 - Port Townsend vessel station 6 (PTV6), benthic infauna sample (B), large van Veen sampler (L), replicate number 1 sieved on a 0.5 mm mesh sieve (15).
- PTV6BL11 - Same as previous example except this sample represents the 1.0-mm sieve size fraction.
- PTV3BS2 - Port Townsend vessel station 3 (PTV3), benthic infauna sample (B), small van Veen samples (S), replicate number 2.

duplex?

The remainder of this report describes departures from the sampling and analysis plan and general observations made in the field.

DEPARTURES FROM THE SAMPLING PLAN

A few departures were made from the sampling and analysis plan. Most of the departures were related to the minor relocations of sampling stations that resulted from instructions conveyed by the EPA lead investigator as sampling proceeded. The other departures from the sampling and analysis plan were that

TABLE 1. SUMMARY OF SAMPLING ACTIVITIES
FOR SALMON NET-PEN STUDY

Date ^a	Crew	Station Sampled ^b	Variables Sampled ^c	Departed Dock	Arrived Dock
May 1	Chip Hogue, PTI Jane Sexton, PTI Kris Flint, EPA Ann Dailey, EPA	PTV1, reference	C,B,W ^d ,P	0900	1630
May 2	Chip Hogue, PTI Jane Sexton, PTI Burney Hill, EPA Lisa Macchio, EPA	PTV2, PTV3, PTV4 ^e	C,B,P	0830	1900
May 3	Chip Hogue, PTI Jane Sexton, PTI Burney Hill, EPA	PTV4 ^f , PTV5, PTV6	C,B,W ^d ,P	0830	1900

^a All dates are 1991.

^b Fish pen location is shown in Figure 1.

^c C - sediment chemistry

B - benthic infauna

P - phospholipid

W - water quality.

^d Water quality samples collected at Stations PTV1 and PTV6.

^e Only 0.1 m² van Veen samples collected.

^f Only 0.025 m² van Veen samples collected.

TABLE 2. SUMMARY OF SAMPLES COLLECTED
DURING THE SALMON NET-PEN STUDY

Station	Distance from Fish-Pens (feet)	Sample Type ^a	Number of Samples ^b	Sample Status
PTV1	1,000	C	5	analyze
		B	10	analyze
		P	3	analyze
		W	1	analyze
PTV2	300	C	5	analyze
		B	10	analyze
		P	3	archive
PTV3	200	C	5	analyze
		B	10	analyze
		P	3	archive
PTV4	100	C	5	analyze
		B	10	analyze
		P	3	archive
PTV5	60	C	5	analyze
		B	10	analyze
		P	3	analyze
		W	1	analyze
PTV6	10	C	5	analyze
		B	10	analyze
		P	3	analyze
		W	1	analyze
Reference	--	C	5	analyze
		B	10	analyze
		P	1	analyze
		P	2	archive

^a C - sediment chemistry
 B - benthic infauna
 P - phospholipid
 W - water quality.

^b Five benthic infauna samples were collected with a 0.1 m² van Veen sampler and five benthic infauna samples were collected with a 0.25 m² van Veen sampler.

TABLE 3. SUMMARY OF STATION CHARACTERISTICS

Station	Loran-C Coordinates	North Latitude	West Longitude	Approx. Distance from Fish-Pens (feet)
PTV1	28342.9 42278.6	48°06.18'	122°46.39'	1,000
PTV2	28343.1 42278.0	48°06.14'	122°46.54'	300
PTV3	28343.1 42278.0	48°06.13'	122°46.55'	200
PTV4	28343.1 42277.8	48°06.13'	122°46.58'	100
PTV5	28343.1 42277.8	48°06.12'	122°46.59'	60
PTV6	28343.1 42277.8	48°06.12'	122°46.60'	10
Reference	28333.6 42275.6	48°05.05'	122°46.59'	--

bacteria samples were collected at all the stations and water samples were collected at two stations.

FIELD OBSERVATIONS

The following notable observations were made during the cruise:

■ Specific comments

- **Station PTV1**—All grabs were full and the sediment was within 1 cm of the top of the grab sampler. The sediment appeared to be brown clay with no odor. Water temperature (1 meter off the bottom) at 1425 on May 3, 1991 (water bottle sample) was 11°C and dissolved oxygen was 7.5 mg/L.
- **Station PTV2**—All grabs were full and the sediment was within 1 cm of the top of the grab sampler. The sediment was medium brown mud and had no odor.
- **Station PTV3**—All grabs were full and the sediment was within 1 cm of the top of the grab sampler. The sediment was medium brown in color. (odor?)
- **Station PTV4**—All grabs were full and the sediment was within 1 cm of the top of the grab sampler. There appeared to be fewer animals present in this sample compared to stations PTV1, PTV2, PTV3, and the reference station. Approximately 100 grams of wood chips were retained in both the 0.1-m² and 0.05-m² sieves. There were more wood chips in this sample than at the other stations. The mud was approximately 10 cm deep and had a black upper layer. (odor?)
- **Station PTV5**—All grabs were full and the sediment was within 1 cm of the top of the grab sampler. Benthic replicate samples numbers 1-4 had 2 cm of brown material on the surface. Sample Number 5 had bacteria on the surface. All of the samples collected contained black mud and had a hydrogen sulfide odor.
- **Station PTV6**—All grabs were full and the sediment was within 1 cm of the top of the grab sampler. Sediments were black with strong hydrogen sulfide odor. The water sample was collected 1 meter from the bottom at 1415 on May 3, 1991. The water temperature and the dissolved oxygen were measured onboard the vessel. The measurements were 11°C and 8.0 mg/L, respectively.

- **Reference station**—All grabs were full and the sediment was within 1 cm of the top of the grab sampler. The sediment appeared to be brown clay with no odor. There appeared to be fewer animals at the reference station than at PTV1, but the sediment that was collected appeared to be almost identical to that at PTV1.
- **General observations**
 - Station positioning or relocation appeared to be very accurate. The accuracy for repeated sampling at a station was usually within 3 meters of the previous sample.
 - The abundances of benthic infauna at Stations PTV5 and PTV6 appeared to be low relative to all other stations sampled.

3.0 CRUISE REPORTS DIVER CORE SAMPLING

On board Chemistry

EPA divers collected sediment and infauna samples at five salmon net-pen facilities: ScanAm #1 (Cypress Island, near Anacortes), Sea Farms WA #1 (Port Angeles), Global Aqua #2 (Rich Passage, near Bainbridge Island), and Global Aqua #3 (Clam Bay, near Manchester), and Paradise Bay Seafarms (Port Townsend). The divers collected the sediment in 0.01-m² core tubes. PTI assisted in this field effort by supplying a field technician who helped EPA sieve (mesh size = 0.5 mm) and preserve all benthic infauna samples collected by the dive team. Station positions were selected in the field by the EPA lead investigator. A total of 191 ^{cores} at 32 stations were collected during the diver survey. A summary of the samples collected at each station is presented in Table 4. The field observations made on each diver-collected sample are included in Appendix B.

Sample identifiers used for diver collected data are based on a site designator followed by a station number. Sample CLAM3, for example, represents the sample taken at Global Aqua #3 (Clam Bay, near Manchester) Station No. 3.

Additional identifiers were used in the case of Paradise Bay Seafarms (Port Townsend) to distinguish between the diver and vessel surveys performed at that location. Paradise Bay Seafarms' samples were tagged with cruise and sample type in addition to the station and site designators. Station PTD6, for example, was the sample collected at Station Number 6 during the Paradise Bay Seafarms diver survey.

TABLE 4. SUMMARY OF SAMPLES COLLECTED
DURING THE DIVER SAMPLING EVENTS

Sample Type ^a	Number of Samples ^b	Sample Status
S	1	analyze
C	2	analyze
B	3	analyze

- ^a S - sulfide
- C - sediment chemistry
- B - benthic infauna.

- ^b Sediment was collected in 0.01-m² core tubes.

4.0 SEDIMENT AND WATER CHEMISTRY QUALITY ASSURANCE AND QUALITY CONTROL REVIEW

This section documents the results of a quality assurance review of data for conventional analyses of 67 sediment and 2 water samples from Puget Sound salmon net-pen areas. The conventional analyses include determination of total sulfides, ammonia, total Kjeldahl nitrogen (TKN), total phosphorus, biochemical oxygen demand (BOD), chemical oxygen demand (COD), total organic carbon (TOC), grain size distribution, total solids, miscellaneous nutrients (e.g., nitrate, nitrite), settleable solids, total volatile solids (TVS), total suspended solids, and turbidity. This quality assurance report is provided in support of the quality assurance project plan (QAPP) for the salmon net-pens monitoring project (PTI 1991), which addressed the overall data quality objectives (DQOs) for this project. These DQOs are outlined in Table 5.

Specific quality control QC measures were utilized by the laboratory to ensure that the overall project DQOs for precision, accuracy, comparability, representativeness, and completeness were achieved. These measures included the use of standard methods, adherence to established sample handling procedures and holding times, and the analysis of replicates and control samples. These control samples included method blanks, initial and continuing calibration verification standards, and laboratory control samples. A list of quality control measures, their frequency, and their control limits is given in Table 6.

Quality control check samples reveal a great deal about sampling technique, analyst technique, instrument capability, possible sources of contamination, and difficulties with the matrix. When considered as a whole, these pieces of information allow one to make a determination as to what degree the analytical results are useable. The following quality control check samples were used. Table 7 outlines the control measures, frequency, and control limits.

- Initial calibration verification (ICV) standards were run after each calibration of an instrument to verify that the instrument is operating properly, and that the standards are accurate. An ICV is an independent reference standard made from a source different than that of the calibration standards.
- Continuing calibration verification (CCV) standards were run routinely (e.g., every 10 samples, every 2 hours) to regularly verify the ongoing calibration of the analytical system.

TABLE 5. DATA QUALIFIER CODES

Qualifier Code	Description
E	Estimate
G	Estimate is greater than value shown
M	Value is a mean
Q	Questionable value
U	Undetected at the detection limit shown
UE	Detection limit shown is an estimate

Table of all samples: analytes
 to provide overview and
 basis for understanding/
 interpreting discussion under
 following times.

TABLE 6. MINIMUM DATA QUALITY OBJECTIVES

Variable	Matrix	Units	Detection Limit	Analytical			Completeness	Method, Reference	Method Description	Holding Time ^a
				Bias	Precision	Detection Limit				
Benthic macroinvertebrates	sediment	numbers of individuals	N/A	± 5% ^b	NA	95%	Taxonomy to species level PSEP (1986)	Sorting and identification	NA	
Bacterial biomass	sediment		N/A	± 5%	± 10%	95%	White et al. (1979)	Phospholipid	NA	
Biochemical oxygen demand	sediment aqueous	mg/kg mg/L	1,200 4	± 20% ± 20%	± 35% ± 35%	95% 95%	PSEP (1986) 405.1, U.S. EPA (1979)	5 days at 20°C	7 days 48 hours	
Chemical oxygen demand	sediment	mg/kg	500	± 20%	± 35%	95%	PSEP (1986)	Titrimetric	7 days	
Total organic carbon	sediment	percent dry weight	1	± 20%	± 35%	95%	PSEP (1986)	Combustion at 950°C	28 days	
Nitrogen, nitrate and nitrite	aqueous	mg/L	0.2	± 20%	± 35%	95%	353.2, U.S. EPA (1979)	Spectrophotometric, cadmium reduction	28 days	
Nitrogen, Kjeldahl	sediment aqueous	mg/kg mg/L	1 0.1	± 20% ± 20%	± 35% ± 35%	95% 95%	Mod. 351.4, U.S. EPA (1979) 351.4, U.S. EPA (1979)	Potentiometric, ion selective electrode	28 days 28 days	
Nitrogen, Ammonia	sediment aqueous	mg/kg mg/L	0.2 0.05	± 20% ± 20%	± 35% ± 35%	95% 95%	Mod. 350.3, U.S. EPA/OSU ^c 350.3, U.S. EPA (1979)	Potentiometric, ion selective electrode	28 days 28 days	
Nitrogen, total organic	sediment aqueous	mg/kg mg/L	0.8 0.1	± 20% ± 20%	± 35% ± 35%	95% 95%	Mod. 351.4-350.3, U.S. EPA/OSU ^c 351.4-350.3, U.S. EPA (1979)	Kjeldahl minus ammonia nitrogen	28 days 28 days	
Total phosphorus	sediment aqueous	mg/kg mg/L	1 0.01	± 20% ± 20%	± 35% ± 35%	95% 95%	Mod. 365.3, U.S. EPA (1979) 365.3, U.S. EPA (1979)	Colorimetric, ascorbic acid	7 days 28 days	
Total sulfides	sediment	mg/kg	20	± 20%	± 35%	95%	PSEP (1986)	Titrimetric, iodine	7 days	
Grain size	sediment	g dry weight	NA	± 20%	± 35%	95%	PSEP (1986)	Wet seive	6 months	
Total volatile solids	sediment	percent	0.1	± 20%	± 35%	95%	PSEP (1986)	Gravimetric, ignition at 550°C	28 days	
Total suspended solids	aqueous	mg/L	5	± 20%	± 35%	95%	160.2, U.S. EPA (1979)	Gravimetric, dried at 103-105°C	7 days	
Total settleable solids	aqueous	mg/L	0.2	± 20%	± 35%	95%	160.5, U.S. EPA (1979)	Volumetric, Imhoff cone	48 hours	

TABLE 6. (Continued)

Variable	Matrix	Units	Analytical				Method, Reference	Method Description	Holding Time ^a
			Detection Limit	Bias	Precision	Completeness			
Turbidity	aqueous	turbidity units	1	20%	± 35%	95%	180.1, U.S. EPA (1979)	Nephelometric	48 hours
pH	aqueous	pH units	NA	NA	± 0.2 pH units	95%	150.1, U.S. EPA (1979)	Electrometric	analyze immediately

^a Based on both a minimum sorting efficiency and minimum taxonomic identification accuracy of 95 percent.

^b The holding times are calculated from time of sampling.

^c Source: Berg and Gardner 1978

TABLE 7. QUALITY CONTROL MEASURES FREQUENCY,
AND CONTROL LIMITS, CONVENTIONAL VARIABLES

Sample Type	Frequency	Control Limit
Method blank	One per analytical batch or per 20 samples of similar matrix, whichever is more frequent	\leq detection limit
Analytical duplicate	One per analytical batch or per 20 samples of similar matrix, whichever is more frequent	\leq 35 relative percent difference
Initial calibration verification	Once for each time instrument is calibrated	90 - 110 percent
Continuing calibration verification	One for every 10 analyses and following the last samples to be analyzed	90 - 110 percent
Laboratory control sample	One per analytical batch or per 20 samples of similar matrix	80 - 120 percent, or EPA advisory limits
Performance evaluation samples	As required for state accreditation	As determined by interlaboratory precision and bias

- Analytical blanks
 - Method blanks were used during sample analyses to evaluate possible sources of laboratory contamination during the analytical procedure. It is carried through the entire procedure using the same reagents, surrogates, etc., as the samples. Method blanks were prepared at the time of sample preparation for each analytical batch of samples using deionized/distilled water.
 - Calibration blanks were used, when appropriate, to "zero" the instrument. The calibration blank is a sample of laboratory water or solvent containing the same reagents at the same concentration as the calibration standards.
- Field replicate analyses were used to assess the overall precision of the investigation. Replicate samples were collected as separate grab samples at each of the locations to be sampled for sediment analysis.
- Laboratory duplicates were used in order to determine the precision of the inorganic analytical method where matrix spike duplicates are not appropriate.
- Laboratory control samples (LCSs) are reference materials that were used, where available, to provide a further evaluation of laboratory accuracy. These LCS are analyzed using the same sample preparation, reagents, and analytical methods employed for samples. Reference materials were obtained from EPA or another well-documented source.

All of the conventional analyses were performed by Columbia Analytical Services in Kelso, Washington. The quality assurance review included examination and validation of the following laboratory data:

- Sample preparation logs and laboratory worksheets
- All instrument printouts
- Instrument calibration and calibration verification procedures and results
- Sample holding times and chain-of-custody records
- Manual data transcriptions.

Data qualifiers are notations that are used by data reviewers to briefly describe or qualify data and the systems producing data. When assigned to individual data points they provide additional information on how, and to what extent, the different QA issues apply to various analytes. Data qualifiers were assigned as necessary during the quality assurance review. Following the validation procedures, data quality was assessed with respect to accuracy, precision, and completeness. All qualifier codes used in this report are defined in Table 5. Sample results, with qualifiers, and summaries of analytical and field precision results are provided in Appendix C. In addition to summarizing the data, the data table in Appendix C provides an integrative presentation of the impact of QA/QC shortfalls on different analytes.

Holding time summaries are presented in Appendix D. Chain-of-custody records are reproduced in Appendix F.

OVERALL CASE ASSESSMENT

Why not → This QA/QC review encompassed 1,162 data points. Grain size and total solids determinations constituted 58.6 percent of this total, and none of these data were qualified. Of the remaining conventional analytes, 5.4 percent were qualified as estimates (*E*), 6.9 percent were qualified as minimum estimates (*G*), 6.4 percent were qualified as undetected (*U*) at the corresponding reporting limit, 1.2 percent were qualified as undetected at an estimated detection limit (*UE*), and 0.4 percent (2 data points) were qualified as questionable values (*Q*). The two data points qualified as questionable were BOD samples (PTV3C4 and PTV3C5), two of five replicate samples.

Data qualified as estimates (*E*), minimum estimates (*G*), or undetected at an estimated detection limit (*UE*) are acceptable, but a greater degree of uncertainty is associated with these values than with unqualified data.

COMPLETENESS

Complete data packages were submitted by Columbia Analytical Services for 67 sediment and 2 water samples. The data were reviewed in accordance with requirements of the QAPP and laboratory statement of work (SOW) for this project (PTI 1991). Because of a change in sampling events, several events that were originally planned to be independent were completed concurrently.

HOLDING TIMES

Holding times specified in the QAPP were met for TOC, TVS, total suspended solids, and nitrate and nitrite.

Analyses of nine BOD samples exceeded the 7-day holding time requirement. Six BOD analyses exceeded the holding time by 1 day. *One analysis exceeded the holding time by 6 days. Results for these eight analyses were qualified as estimates (E). Analyses of two samples (two of five replicates) exceeded the holding time by 33 days because the samples were temporarily lost by the laboratory. Results for these samples were qualified as questionable values (Q).

The contract with the laboratory specified the PSEP-recommended holding times. In many instances, the laboratory did not meet these recommendations.* In these instances, additional information was considered, and other precedents were reviewed to determine if the data could still be considered valid, without qualification. The largest body of research pertaining to holding times is that which was performed to validate aqueous holding times. When reviewing the data for this project, these holding times were considered and were used to assist in the assignment of qualifiers.

Analyses of 11 sulfide samples did not meet the 7-day holding time requirement. Because of this and other quality control exceedances, the associated results were qualified as minimum estimates (G) and undetected values were qualified as estimated detection limits (UE).

Analyses of seven samples did not meet the 28-day holding time requirement for total organic nitrogen (TON). Results for these samples were qualified as estimates (E).

None of the sediment samples were analyzed for total phosphorus within the SOW holding time of 7 days. However, most analyses did meet the 28-day holding time recommended for wastewater analyses (U.S. EPA 1983). The total phosphorus data were not qualified if this 28-day holding time was met. Analyses of seven samples exceeded the latter holding times, and the associated results were qualified as estimates (E).

Twenty-two sediment samples were not analyzed for COD within the SOW holding time of 7 days. However, all of these samples were analyzed within the 28-day holding time recommended for wastewater (U.S. EPA 1983). Therefore, the data were not qualified.

Both water samples were analyzed for ammonia and TKN past the recommended holding time of 28 days. Results for these analyses were qualified as estimates (E).

The water samples to be analyzed for settleable solids and turbidity were received at the laboratory past the 48-hour holding time requirement. The laboratory analyzed the samples upon receipt. Because of the possibility of decomposition, the results for these analyses were qualified as estimates (E) or as having estimated detection limits (UE).

ANALYTICAL METHODS AND QUALITY ASSURANCE REVIEW

All sample extraction and analysis procedures, instrument calibration procedures, and quality control checks conformed to QAPP requirements, except as discussed in the following sections.

Total Volatile Solids

Sediment samples were analyzed for TVS by the Puget Sound Estuary Program (PSEP) method (PSEP 1986). Percent TVS represent the fraction of total solids that are lost on ignition at a temperature that is sufficient to vaporize organic material. Therefore, TVS can be used as an approximate indicator of the amount of organic matter in the total solids (PSEP 1986), although some inorganic material is also vaporized in the procedure.

The sample, which was dried at $103 \pm 2^\circ\text{C}$ for total solids determinations, was ignited at $550 \pm 10^\circ\text{C}$ to a constant weight, cooled in a desiccator, and then weighed. The portion of solids which is lost upon ignition is the percent TVS.

Accuracy—No laboratory control samples (LCSs) were analyzed for TVS with these samples. This omission is acceptable. The low-level LCSs, available commercially are inappropriate for sediment TVS determinations. There were no other available reference standards that may have been used for the determination of TVS in sediment samples.

The analytical balance calibration was verified on each day of use with S-class weights. The drying oven thermometer was not calibrated against a standardized thermometer approved by the National Institute for Standards and Testing, but the oven temperature was monitored on each day using a commercial thermometer. The use of a non-verified commercial thermometer generally introduces an uncertainty of $\pm 5^\circ\text{C}$, but it is not a cause for concern. There are no other controls to be placed on this analysis by the laboratory.

Precision—Triplicate analyses were performed once for every analytical batch of twenty or fewer samples. Replicate field samples were taken at 5 stations and the reference area. The precision, in percent relative standard deviation (RSD), in all cases met the performance criterion of the study, except in the set PTV1C, which yielded a precision of 56 percent RSD. One of the replicate samples of the group was considered to be an outlier; recalculation without this sample yields 1.8 percent RSD.

Wrong method?

Total Sulfide

Sediment samples were analyzed for total sulfide by the PSEP/9030 method (PSEP 1986). Total sulfides represent the amount of acid-soluble hydrogen sulfide, HS^- , and S^{2-} in a sample. Sulfides are measured because they may be toxic and may create unaesthetic conditions (PSEP 1986).

Excess iodine was added to a sample which had been treated with zinc acetate to produce zinc sulfide. The iodine oxidizes the sulfide to sulfur under acidic conditions. The excess iodine was then backtitrated with sodium thiosulfate.

Accuracy—The accuracy of the titration was verified through the analysis of an LCS of sodium sulfide. In addition, a sample from each analytical batch was spiked with a known amount of sulfide. Samples were then analyzed. An LCS was analyzed after every 10 samples and following the last analysis.

The effect of matrix interferences on accuracy was assessed through the use of matrix spikes, for which no quality control criteria were specified in the QAPP. The percent recoveries of these matrix spikes ranged from 55 to 98 percent. The implied recovery uncertainty of up to a factor of 2 was acceptable for sediment matrix.

Precision—Duplicate analyses were performed once for every 20 or fewer samples in an analytical batch. Replicate field samples were taken at five stations and the reference area.

Sulfide is volatilized by aeration and any oxygen inadvertently added to the sample may have converted the sulfide to an unmeasurable form. There was a high percent RSD between field samples, probably attributable to aeration in the field or in the laboratory. Therefore, all measurements were considered minimum estimate, and were qualified with a *G* qualifier.

Total Organic Nitrogen

Sediment samples were analyzed for TON through the analysis of TKN and ammonia. These analyses were conducted by revised EPA Methods 351.4 and 350.3, respectively (U.S. EPA 1983). TKN is the sum of ammonia plus TON. Therefore, TON is TKN minus ammonia.

Ammonia (Sediment)—Sediment samples were analyzed for ammonia using a method developed by Oregon State University (Berg and Gardner 1978). An aliquot of sample was extracted with 2M potassium chloride. The extract was

then brought to a known volume and analyzed as an aqueous sample according to EPA Method 350.3 (U.S. EPA 1983). Ammonia in the extract was determined potentiometrically using an ion-selective ammonia electrode and a specific ion meter. The ammonia electrode uses a hydrophobic gas-permeable membrane to separate the sample solution from a solution of ammonium chloride. Ammonia in the sample diffuses through the membrane and alters the pH of the ammonium chloride solution, which is sensed by a pH electrode. The constant level of chloride in the ammonium solution is sensed by a chloride-selective ion electrode, which acts as the reference electrode (U.S. EPA 1983).

Total Kjeldahl Nitrogen (Sediment)—An aliquot of each sediment sample was digested with sulfuric acid for TKN analysis. The digestate was then brought to a known volume and was analyzed as an aqueous sample according to EPA Method 351.4 (U.S. EPA 1983). Following digestion and cooling, distilled water was added to the digestion flask and the pH was adjusted to between 3 and 4.5 by the addition sodium hydroxide. The sample was then cooled and transferred to a 100 mL beaker. After inserting the electrode into the sample, a solution of sodium hydroxide, sodium iodide, and ethylenediaminetetraacetic acid (EDTA) was added and the ammonia measured potentiometrically.

Accuracy—The TKN instrument was calibrated using a blank and four standards. The calibration was then verified with an initial calibration verification (ICV) standard obtained from a commercial source. The concentration of the ICV was verified by the analysis of a continuing calibration verification (CCV) standard. Samples were then analyzed. After every 10 samples, and following the last analysis, a CCV and a continuing calibration blank (CCB) were analyzed.

The laboratory used the ICV standard as an LCS, although the standard was not digested with the samples. Therefore, the laboratory used an 85–115 percent control window for LCS analyses, which exceeded the 95–100 percent control window for ICV standards on several occasions. The results were not qualified for this discrepancy. The CCV standard recoveries were within the 90–110 percent control windows.

Precision—Triplicate analyses were performed once for every 20 or fewer samples in an analytical batch. Replicate field samples were taken at five stations and the reference area. The results, in percent RSD, were well within the goals of the study for analytical precision. However, field replicate precision was very poor for three of the areas tested (PTV1C, PTV2C, and PTV5C). The percent RSD yielded 61, 62, and 70 percent, respectively. None of these instances can be conclusively called an outlier, and the source of variability could not be determined.

Total Phosphorus

Sediment samples were analyzed for total phosphorus by EPA Method 365.3 (U.S. EPA 1983). Ammonium molybdate and antimony potassium tartrate were reacted in an acid medium with dilute solutions of phosphorus to form an antimony-phospho-molybdate complex. This complex was reduced to an intensely blue-colored complex by addition of ascorbic acid. The color is proportional to the phosphorus concentration and was measured spectrometrically.

Accuracy—The total phosphorus instrument was calibrated using a blank and five standards. The calibration was then verified with an ICV standard made from an EPA quality control check sample. An initial calibration blank (ICB) was also analyzed. In all cases the ICV and ICB control limits (90-110 percent and less than the detection limit, respectively) were met. The concentration of the ICV standard was verified by the analysis of a CCV standard obtained from a commercial source. Samples were then analyzed. After every 10 samples, and following the last, analysis a CCV and a CCB were analyzed. In all cases the control windows for the CCVs and CCBs were met.

Precision—Triplicate analyses were performed once for every analytical batch of twenty or fewer samples. The analytical precision, in percent RSD, met the study criteria of ± 35 percent RSD.

Five field replicates were taken from five stations and a reference area. The field precision calculated for these replicate samples met the study criterion in all cases (± 35 percent RSD).

Biochemical Oxygen Demand

Sediment samples were analyzed for BOD by the PSEP (1986) method. An aliquot of sample was weighted and transferred to a BOD bottle. Dilution water was added, making sure that no air bubbles were trapped in the bottle. The initial dissolved oxygen concentration was determined, then the samples were incubated for 5 days at $20 \pm 1^\circ\text{C}$. A dissolved oxygen concentration was again determined. BOD is the measure of the dissolved oxygen consumed by microbial organisms while assimilating and oxidizing the organic matter in a sample. This test is used to estimate the amount of organic matter that is available to organisms, in contrast to other tests used to estimate the total amount of organic matter. In addition to oxygen used for degrading organic matter, BOD may also include oxygen used to oxidize inorganic material and reduced forms of nitrogen (PSEP 1986).

Accuracy—Accuracy of the method was established by the analysis of an LCS of glucose-glutamic acid with each analytical batch of 20 or fewer samples. Results for all LCS standards analyzed were within the LCS control window of 80-120 percent.

Precision—Triplicate analyses were performed once for every 20 or fewer samples in an analytical batch. Replicate field samples were taken at five stations and the reference area. The results, in percent RSD, are within the goals of the study of ± 35 percent RSD, with the exception of the PTV2C field replicates. In this instance, one of the five samples was considered to be an outlier (Crow et al 1960). The recalculation of percent RSD, excluding this result, yields a value of 18.6 percent, which is within the control window. Therefore, no qualification of the data was made based upon precision data.

Chemical Oxygen Demand

Sediment samples were analyzed for COD by the PSEP (1986) method. COD is a measure of the oxygen equivalent of the organic matter content of a sample that is susceptible to oxidation by a strong chemical oxidant at elevated temperature and reduced pH. The test was devised as an alternative to the BOD test for estimating organic matter. For samples from a specific source, COD can be related empirically to BOD, TOC, or TVS and then used for monitoring after a relationship has been established.

Major limitations of the COD test are that it is not specific for organic matter and that correlations with other measures of organic carbon are not always found. Inorganic substances such as Fe^{+2} , Mn^{+2} , and S^{-2} can increase the consumption of oxidizing agent during the test. Plumb (1981) recommends that COD not be equated with organic matter in sediments (PSEP 1986).

The samples were warmed to room temperature, homogenized, and a representative aliquot was taken and weighed. The aliquot was then quantitatively transferred to a COD reflux flask, along with mercuric sulfate and potassium dichromate. Sulfuric acid-silver sulfate solution was added, and the mixture was refluxed for 2 hours, then cooled. Ferroin indicator was added and the sample was titrated with ferrous ammonium sulfate.

Accuracy—The accuracy of the titration was verified through the analysis of an LCS of glucose. The LCS was analyzed once per analytical batch of 20 or fewer samples. The LCS analyses met the 90-110 percent control limit. In addition, method blanks were analyzed at the same frequency, and COD was not detected in these blanks.

Precision—Triplicate analyses were performed once for every analytical batch of 20 or fewer samples. The analytical precision, in percent RSD, met the study criterion of ± 35 percent RSD.

Five field replicates were taken from five stations and a reference area. The field precision calculated for these replicate samples met the study criterion in all cases (± 35 percent RSD).

Total Organic Carbon

Sediment samples were analyzed for TOC by the PSEP (1986) method. TOC is a measure of the total amount of nonvolatile, volatile, partially volatile, and particulate organic compounds in a sample. TOC is independent of the oxidation state of the organic compounds and is not a measure of the organically bound and inorganic elements that can contribute to the BOD and COD tests (PSEP 1986).

Samples were dried to a constant weight at a temperature of $70 \pm 2^\circ\text{C}$. The drying temperature is relatively low to minimize loss of volatile organic compounds. After cooling in a desiccator, the sample was ground by mortar and pestle to break up aggregates. A representative aliquot was transferred to a clean, preweighed combustion boat and weighed. Carbonates were removed from the sample by the addition of hydrochloric acid, then the sample was dried again at $70 \pm 2^\circ\text{C}$. Previously ashed cupric oxide fines were then added to the combustion boat, and the samples were combusted at a temperature of $950 \pm 10^\circ\text{C}$, to yield carbon dioxide, which was measured coulometrically.

Accuracy—The TOC instrument was calibrated according to the manufacturer's directions. The calibration was then verified with an ICV standard of (EPA) municipal digested sludge. The instrument baseline was determined by the analysis of an ICB. The concentration of the ICV was verified by the analysis of a CCV standard of urea. Samples were then analyzed. After every 10 samples, and following the last analysis, a CCV and a CCB were analyzed.

ICV percent recovery was found to be within the 80-120 percent control window. The ICB and CCB were found to be less than the detection limit. The CCV percent recovery was found to be within the 90-110 percent control window.

Precision—Triplicate analyses were performed once for every 20 or fewer samples in an analytical batch. Replicate field samples were taken at five stations and the reference area. The results, in percent RSD, were well within the goals of the study (± 35 percent RSD).

Grain Size and Total Solids

Sediment samples were analyzed for particle size determination by the PSEP (1986) method. Particle size is used to characterize the physical characteristics of sediments. Because particle size influences both chemical and biological variables, it can be used to normalize chemical concentrations according to sediment characteristics and to account for some of the variability found in biological assemblages (PSEP 1986).

Samples were homogenized and a representative subsample of approximately 10 grams (wet weight) was removed. Total solids (percent) was determined by drying the aliquot to a constant weight at a temperature of $103 \pm 2^\circ\text{C}$, cooling to room temperature in a desiccator, then weighing the cooled sample.

A second representative subsample of approximately 30 grams (wet weight) was taken for wet sieving. Wet sieving separates the sample into size fractions greater than $62.5 \mu\text{m}$ (i.e., sand and gravel) and less than $62.5 \mu\text{m}$ (i.e., silt and clay). The sand and gravel fraction was subdivided further by mechanically dry sieving it through a graded series of screens. The silt-clay fraction was subdivided further using a pipet technique that depends upon the differential settling rates of different sized particles.

Precision—Triplicate analyses were performed once for every analytical batch of 20 or fewer samples. The analytical precision, in percent RSD, met the study criterion of ± 35 percent RSD except in two cases (detailed below). For grain size, this criteria was not used when the size fraction constituted less than 10 percent of the total mass because in these small fractions there is greater variability.

For grain size, the analysis of the PTDC set of analytical triplicate samples showed two of the samples to be comparable, while the third (composed primarily of clay) differed in its grain size distribution. Therefore, the percent RSDs for this analytical batch were high. Likewise, the field precision of total solids measurements for the PTDC set was poor. Perhaps because of the high clay content mixing in the laboratory was not effective. It is recommended that averaging the PTDC replicate values be considered during data analysis to account for this heterogeneity. For grain size, the analysis of analytical triplicate samples Station PTVC3C) showed a percent RSD between the slit and clay fraction measurements of 38 percent and 87 percent, respectively.

Five field replicates were taken from five stations and a reference area. For grain size, the analytical precision for fractions that comprised at least 10 percent of the sample was good, within ± 35 percent except for the PTVC (62 percent RSD) and REFCO (38 percent RSD) between replicate factions.

Accuracy—No LCSs were analyzed with these samples for percent solids. This omission is acceptable. The low-level LCSs available commercially are inappropriate for sediment percent solids determinations. There were no other available reference standards that could be used for the determination of total solids in sediment samples.

The analytical balance calibration was verified on each day of use with S-Class weights. The drying oven thermometer was not calibrated against a standardized thermometer approved by the National Institute for Standards and Testing, but the oven temperature was monitored on each day of use by a commercial thermometer and was recorded. The use of a non-verified commercial thermometer generally introduces an uncertainty of $\pm 2^{\circ}\text{C}$, but it is not a cause for concern. As the procedure for determining particle size is a mechanical procedure, there are no other controls to be placed on this experiment by the laboratory.

Ammonia (Water)

Water samples were analyzed for ammonia by EPA Method 350.3 (U.S. EPA 1983). Sample concentrations were determined potentiometrically using an ion selective ammonia electrode and a specific ion meter. The ammonia electrode uses a hydrophobic gas-permeable membrane to separate the sample solution from an ammonium chloride solution. Ammonia in the sample diffuses through the membrane and alters the pH of the ammonium chloride solution, which is sensed by a pH electrode. The constant level of chloride in the ammonium chloride solution is sensed by a chloride selective electrode, which acts as the reference electrode (U.S. EPA 1983).

Accuracy—The instrument was calibrated using a blank and four standards. The calibration was then verified with an LCS obtained from a commercial source. The concentration of the LCS was verified by the analysis, of a CCV standard. Samples were then analyzed. After every 10 samples, and following the last analysis, a CCV and a CCB were analyzed. The LCS was within the control limit window of 80-120 percent variation. The CCV was within the control limit window of 90-110 percent variation. There was nothing detected in the method blank or the CCB.

Precision—Triplicate analyses were performed once per analytical batch of 20 or fewer samples. The analytical precision met the percent RSD criterion of ± 35 percent RSD.

Nitrate and Nitrite

Water samples were analyzed for nitrate and nitrite by EPA Method 353.2 (U.S. EPA 1983). The sample was passed through a column containing granulated copper-cadmium to reduce the nitrate to nitrite. The nitrite was determined by diazotizing with sulfanilamide and coupling with N-(1-naphthyl)-ethylenediamine dihydrochloride to form a highly colored azo dye, which was measured calorimetrically.

Accuracy—The nutrient analyzer was calibrated using a blank and four standards. The calibration was then verified with an LCS obtained from a commercial source. The concentration of the LCS was verified by the analysis of a CCV standard. Samples were then analyzed. After every 10 samples, and following the last analysis a CCV and a CCB were analyzed. The LCS was within the control limit window of 80-120 percent variability. The CCV was within the control limit window of 90-110 percent variability. There was nothing detected in the method blank or the CCB.

Precision—Duplicate analyses were performed once per analytical batch of 20 or fewer samples. The analytical precision met the percent RSD criterion of ± 35 percent RSD.

Total Kjeldahl Nitrogen (Water)

Water samples were analyzed for TKN by EPA Method 351.4 (U.S. EPA 1983). Following digestion, distilled water was added to the digestion flask and the pH was adjusted to between 3 and 4.5 by the addition of sodium hydroxide. After inserting the electrode into the sample, sodium hydroxide, sodium iodide, and EDTA was added and the ammonia measured potentiometrically.

Accuracy—The instrument was calibrated using a blank and four standards. The calibration was then verified with an LCS obtained from a commercial source. The concentration of the LCS was verified by the analysis of a CCV standard. Samples were then analyzed. After every 10 samples, and following the last analysis, a CCV and a CCB were analyzed. The LCS was within the control limit window of 80-120 percent variability. The CCV was within the

control limit window of 90-110 percent variability. There was nothing detected in the method blank or the CCB.

Precision—Triplicate analyses were performed once per analytical batch of 20 or fewer samples. The analytical precision met the percent RSD criterion of ± 35 percent RSD.

Settleable Solids

Water samples were analyzed according to EPA Method 160.5 (U.S. EPA 1983). The sample was transferred to an Inhoff cone, and matter was allowed to settle. The cone is marked, like a volumetric flask. The amount of material that settles is determined by reading the closest mark to the separation between the aqueous and solid layers.

Accuracy—There were no accuracy controls specified in this analysis, because the laboratory used volumetric glassware to make the determination.

Precision—No analytical precision measurements were made for this analysis.

Total Suspended Solids

Water samples were analyzed for total suspended solids by EPA Method 160.2 (U.S. EPA 1983). Well mixed samples were filtered through a glass fiber filter, and the residue retained on the filter was dried to a constant weight at 103-105°C.

Accuracy—An LCS obtained from a commercial source and a method blank were analyzed with the samples. The LCS recovery was 97 percent of the true value. This is within the study goal of 80-120 percent and indicates very good accuracy and interlaboratory comparability. The method blank was free of contamination.

The analytical balance calibration was verified on each day of use with S-class weights. The drying oven thermometer was not calibrated against a standardized thermometer approved by the National Institute for Standards and Testing, but the oven temperature was monitored and recorded on each day using a commercial thermometer. The use of a non-verified thermometer generally introduces an uncertainty of $\pm 2^\circ\text{C}$, but it is not a cause for concern.

Precision—Triplicate analyses were performed once per analytical batch of 20 or fewer samples. The analytical precision met the percent RSD criterion of ± 35 percent RSD.

Turbidity

Water samples were analyzed for turbidity by EPA Method 180.1 (U.S. EPA 1983). The method is based upon a comparison of the intensity of light scattered by the sample under defined conditions with the intensity of light scattered by a standard reference suspension. The higher the intensity of scattered light, the higher the turbidity. Readings, in nephelometric turbidity units (NTU), were made in a nephelometer. A standard suspension of formazin, prepared under closely defined conditions, was used to calibrate the instrument.

Accuracy—An LCS obtained from a commercial source and a method blank were analyzed with the samples. The LCS recovery was 97 percent. The method blank was free of contamination.

Precision—Triplicate analyses were performed once per analytical batch of 20 or fewer samples. The analytical precision met the percent RSD criteria of ± 35 percent RSD.

5.0 BENTHIC INFAUNA QUALITY ASSURANCE AND QUALITY CONTROL REVIEW

Benthic infauna assemblages were sampled and analyzed at five salmon net-pen sites in Puget Sound in accordance with PSEP protocols. A total of 32 stations were sampled by EPA divers with three replicates collected at each station using core tubes of approximately 10 cm in diameter. All diver collected samples were sieved using a 0.5 mm mesh size.

A vessel was also used to collect two additional types of benthic samples at one of the net-pen sites. At Paradise Bay Seafarms (Port Townsend), the *R/V Kittiwake* was used to collect five replicate 0.025-m² and five replicate 0.1 m²-van Veen grab samples at each of seven stations (six net-pen stations and one reference station). All vessel-collected samples were sieved using a 0.5-mm mesh size. The samples collected with the 0.1 m²-van Veen sampler were also sieved using a 1.0-mm mesh size. This sampling design was used to compare the 0.5-mm mesh size fraction between the two van Veen samplers and to compare the two size fractions (0.5 mm and 1.0 mm) of infauna for the larger van Veen sampler.

All samples were sorted, and individuals were identified to the lowest possible taxonomic level. Data were reviewed for:

- Sorting efficiency
- Taxonomic identifications
- Numerical abundance.

Benthic data were sorted and identified by E.V.S. Consultants, Seattle, Washington.

SORTING EFFICIENCY

Twenty percent of each sample was resorted by a person other than the one who originally sorted the sample. In 6 of 204 cases (3 percent), the number of organisms found during resorting, when corrected for the volume of the sample that was re-sorted, was greater than 5 percent of the total number of organisms in the sample (i.e., sorting efficiency was less than the desired level of 95 percent). Those samples included:

- PTAN2-2

- PTV1-11 (0.5-mm fraction)
- PTV2-25 3
- PTV3-11 (0.5-mm fraction)
- PTV3-14 (0.5-mm fraction)
- PTV3-15 (0.5-mm fraction)

Each of the above samples was completely resorted and subjected to a second 20 percent QA/QC evaluation. The desired sorting efficiency of ≥ 95 percent was achieved for all of these samples after resorting. No samples sieved on a 1.0 mm sieve required resorting.

TAXONOMY

Five percent of each major taxon were sent to recognized taxonomic experts outside of the E.V.S. laboratory. Mr. Howard Jones of Marine Taxonomic Services provided QA/QC for polychaetes, molluscs, and miscellaneous taxa. Arthropods were checked by Ms. Pamela Sparks. For all groups, external QA/QC confirmed taxonomic accuracy within the 95 percent limits required by the PSEP protocols. Those discrepancies noted are discussed below.

Polychaetes

A discrepancy was reported for polychaete identifications. *Prionospio cirrifera* was re-identified as *Prionospio multibranchiata*. The change was incorporated into the final data set.

Arthropods

Original identification of the crab *Pinnixa* indicated resemblance to *P. schmitti*. External review confirmed this species identification and all occurrences of *Pinnixa* cf. *schmitti* were changed to *Pinnixa schmitti*.

Molluscs

A difference in taxa was noted in sample PTV6-11 (0.5-mm fraction). *Turbonilla* spp. and *Nuculana* spp. (juveniles) were identified by external re-identification, but the sample was originally identified as containing *Alvania* cf. *compacta* and *Acila castrensis*. This difference was probably due to the small size and early life stage of the organisms. Subsequent re-examination by EVS of

the original specimens confirmed the original identifications, and no changes were made to the data.

In addition to the external review of species identifications, the data collected at the Port Townsend reference station (PTVREF) were compared to results of previous studies. The Puget Sound Ambient Monitoring program has collected benthic infauna samples at this location previously. Comparison of the species list from the previous study and that of the salmon net-pen sampling indicated general agreement in the species composition at the station.

NUMERICAL ABUNDANCE

Numerical abundances of benthic infauna are typical for what is expected at the stations sampled. However, the coefficient of variation for total infauna abundance was unusually large at seven stations (ANAC-1, ANAC-4, BAIN-1, CLAM-5, MANC-1, PTAN-5, PTV6). This variability is caused by the presence of large numbers of nematodes in some of the replicate core or grab samples. Nematodes are traditionally not sampled quantitatively on 0.5 or 1.0-mm mesh sieves because the majority of individuals pass through these screens. The presence of mucous or wood debris in a sample can occasionally artificially decrease the effective mesh size of a sieve and result in the capture of large numbers of nematodes. Therefore, the variability present within the data is considered to be an artifact of sampling and not the result of laboratory sorting problems. Users of this data should consider excluding nematodes for data analysis.

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