Recommended Protocols for Measuring Conventional Sediment Variables in Puget Sound

For

U.S. Environmental Protection Agency
Region 10, Office of Puget Sound
1200 6th Avenue
Seattle, WA 98101

Puget Sound Water Quality Authority
P. O. Box 40900
Olympia, WA 98504-0900

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CONTENTS

	Page
LIST OF TABLES	v
INTRODUCTION	1
COLLECTION OF SURFICIAL SEDIMENTS FOR PHYSICAL AND CHEMICAL VARIABLES	4
INTRODUCTION	4
DESIGN OF SAMPLER	4
PENETRATION DEPTH	5
OPERATION OF SAMPLER	5
SAMPLE ACCEPTABILITY CRITERIA	7
SAMPLE COLLECTION	7
PARTICLE SIZE	9
USE AND LIMITATIONS	9
FIELD PROCEDURES	9
Collection	9
Processing	10
LABORATORY PROCEDURES	10
Analytical Procedures	10
QA/QC Procedures	15
DATA REPORTING REQUIREMENTS	16
TOTAL SOLIDS	17
USE AND LIMITATIONS	17
FIELD PROCEDURES	17
Collection	17
Processing	17
LABORATORY PROCEDURES	17
Analytical Procedures	17
QA/QC Procedures	18
DATA REPORTING REQUIREMENTS	19

TOTAL VOLATILE SOLIDS (TVS) 20

USE AND LIMITATIONS 20
FIELD PROCEDURES 20

Collection 20
Processing 20

LABORATORY PROCEDURES 20

Analytical Procedures 20
QA/QC Procedures 21

DATA REPORTING REQUIREMENTS 22

TOTAL ORGANIC CARBON (TOC) 23

USE AND LIMITATIONS 23
FIELD PROCEDURES 23

Collection 23
Processing 23

LABORATORY PROCEDURES 23

Analytical Procedures 23
QA/QC Procedures 25

DATA REPORTING REQUIREMENTS 25

OIL AND GREASE (FREON EXTRACTABLE) 26

USE AND LIMITATIONS 26
FIELD PROCEDURES 26

Collection 26
Processing 26

LABORATORY PROCEDURES 26

Analytical Procedures 26
QA/QC Procedures 29

DATA REPORTING REQUIREMENTS 29

TOTAL SULFIDES 30

USE AND LIMITATIONS 30
FIELD PROCEDURES 30

Collection 30
Processing 30

LABORATORY PROCEDURES 30

Analytical Procedures 30
QA/QC Procedures 33

DATA REPORTING REQUIREMENTS 34

TOTAL NITROGEN 35

BIOCHEMICAL OXYGEN DEMAND (BOD) 36

USE AND LIMITATIONS 36
FIELD PROCEDURES 36

Collection 36
Processing 36

LABORATORY PROCEDURES 36

Analytical Procedures 36
QA/QC Procedures 39

DATA REPORTING REQUIREMENTS 39

CHEMICAL OXYGEN DEMAND (COD) 40

USE AND LIMITATIONS 40
FIELD PROCEDURES 40

Collection 40
Processing 40

LABORATORY PROCEDURES 40

Analytical Procedures 40
QA/QC Procedures 42

DATA REPORTING REQUIREMENTS 43

REFERENCES 44

LIST OF TABLES

Number		Page
1	Contributors to the sediment conventional protocols	2
2	Recommended sample sizes, containers, preservation, techniques, and holding times for sediment conventional variables	3
3	Withdrawal times for pipet analysis as a function of particle size and water temperature	14

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INTRODUCTION

Recommended methods for measuring the following conventional sediment variables in Puget Sound are presented in this chapter:

- Particle size
- Total solids
- Total volatile solids
- Total organic carbon
- Oil and grease
- Total sulfides
- Total nitrogen
- Biochemical oxygen demand
- Chemical oxygen demand.

Each method is based on the results of a workshop and written reviews by representatives from most organizations that fund or conduct environmental research in Puget Sound (Table 1). The purpose of developing these recommended protocols is to encourage all Puget Sound investigators conducting monitoring programs, baseline surveys, and intensive investigations to use standardized methods whenever possible. If this goal is achieved, most data collected in Puget Sound should be directly comparable and thereby capable of being integrated into a sound—wide database. Such a database is necessary for developing and maintaining a comprehensive water quality management program for Puget Sound.

Each recommended protocol describes the use and limitations of the respective variable; the field collection and processing methods; and the laboratory analytical, QA/QC, and data reporting procedures. Each recommended analytical procedure was modified from Plumb (1981). The general collection and holding recommendations for each variable are presented in Table 2.

Although the following protocols are recommended for most studies conducted in Puget Sound, departures from these methods may be necessary to meet the special requirements of individual projects. If such departures are made, however, the funding agency or investigator should be aware that the resulting data may not be comparable with most other data of that kind. In some instances, data collected using different methods may be compared if the methods are intercalibrated adequately.

TABLE 1. CONTRIBUTORS TO THE SEDIMENT CONVENTIONAL PROTOCOLS

Name	Organization
John Armstrong[a]	U.S. EPA
Bob Barrick[a]	Tetra Tech, Inc.
Scott Becker[a,b]	Tetra Tech, Inc.
Gordon Bilyard[a]	Tetra Tech, Inc.
Chuck Boatman[a]	URS Engineers
Eric Crecelius[a]	Batelle Northwest
Joe Cummins	U.S. EPA
Bob Dexter[a]	EVS Consultants
George Ditsworth	U.S. EPA
John Downing[a]	Nortec
Bruce Duncan	U.S. EPA
Mark Fugiel[a]	Am Test, Inc.
Arnold Gahler	U.S. EPA
Carolyn Gangmark	U.S. EPA
Roy Jones	U.S. EPA
Dave Kendall	U.S. COE
Shawn Moore[a]	Am Test, Inc.
Gary Mauseth[a]	Nortec
Mike Nelson[a]	Laucks Testing Labs
Ahmad Nevissi[a]	Univ. of Washington
Wally Triol[a]	Parametrix, Inc.
Frank Urabeck	U.S. COE
Steve Vincent[a]	Weyerhauser Company
Fred Weinman[a]	U.S. COE
Julia Wilcox[a]	Tetra Tech, Inc.
Carolyn Wilson	U.S. EPA
Jack Word	Evans-Hamilton, Inc.

[a]Attended the workshop held on June 28, 1985.
[b]Workshop moderator.

TABLE 2. RECOMMENDED SAMPLE SIZES, CONTAINERS, PRESERVATION TECHNIQUES AND HOLDING TIMES FOR SEDIMENT CONVENTIONAL VARIABLES

Variable	Minimum Sample Size (g)[a]	Container[b]	Preservation	Maximum Holding Time
Particle size	100-150[c]	P,G	Cool, 4°C	6 mo[d]
Total Solids	50	P,G	Freeze	6 mo[d]
Total Volatile Solids	50	P,G	Freeze	6 mo[d]
Total Organic Carbon	25	P,G	Freeze	6 mo[d]
Oil and Grease	100	G Only	Cool, 4°C, HCl; Freeze	28 days[d] 6 mo[d]
Total Sulfides	50	P,G	Cool, 4°C, lN zinc acetate	7 days[d]
Total Nitrogen	25	P,G	Freeze	6 mo[d]
Biochemical oxygen demand	50	P,G	Cool, 4°C	7 days[e]
Chemical oxygen demand	50	P,G	Cool, 4°C	7 days[e]

[a] Recommended field sample sizes for one laboratory analysis. If additional laboratory analyses are required (e.g., replicates), the field sample size should be adjusted accordingly.

[b] P = polyethylene, G = glass.

[c] Sandier sediments require larger sample sizes than do muddier sediments.

[d] This is a suggested holding time. No U.S. EPA criteria exist for the preservation of this variable.

[e] This holding time is recommended by Plumb (1981).

COLLECTION OF SURFICIAL SEDIMENTS FOR PHYSICAL AND CHEMICAL VARIABLES

INTRODUCTION

This section describes the protocols required to collect an acceptable subtidal surficial sediment sample for subsequent measurement of physical and chemical variables. This subject has generally been neglected in the past and sampling crews have been given relatively wide latitude in deciding how to collect samples. However, because sample collection procedures influence the results of all subsequent laboratory and data analyses, it is critical that samples be collected using acceptable and standardized techniques.

DESIGN OF SAMPLER

In Puget Sound, the most common sampling device for subtidal surficial sediments is the modified van Veen bottom grab. However, a variety of coring devices is also used. The primary criterion for an adequate sampler is that it consistently collect undisturbed samples to the required depth below the sediment surface without contaminating the samples. An additional criterion is that the sampler can be handled properly on board the survey vessel. An otherwise acceptable sampler may yield inadequate sediment samples if it is too large, heavy, or awkward to be handled properly.

Collection of undisturbed sediment requires that the sampler:

- Create a minimal bow wake when descending
- Form a leakproof seal when the sediment sample is taken
- Prevent winnowing and excessive sample disturbance when ascending
- Allow easy access to the sample surface.

Most modified van Veen grabs have open upper faces that are fitted with rubber flaps. Upon descent, the flaps are forced open to minimize the bow wake, whereas upon ascent, the flaps are forced closed to prevent sample winnowing. Some box corers have solid flaps that are clipped open upon descent and snap shut after the corer is triggered. Although most samplers seal adequately when purchased, the wear and tear of repeated field use eventually reduces this sealing ability. A sampler should therefore be monitored constantly for sample leakage. If unacceptable leakage occurs, the sampler should be repaired or replaced. If a sampler is to be borrowed or leased for a project, its sealing ability should be confirmed prior to sampling. Also, it is prudent to have a backup sampler on board the survey vessel in case the primary sampler begins leaking during a cruise.

The required penetration depth below the sediment surface is a function of the desired sample depth (see Penetration Depth). Generally, it is better to penetrate below the desired sample depth to minimize sample disturbance when the sampling device closes. Penetration

depth of most sampling devices varies with sediment character, and generally is greatest in fine sediments and least in coarse sediments. Sampling devices generally rely upon either gravity or a piston mechanism to penetrate the sediment. In both cases, penetration depth can be modified by adding or subtracting weight from the samplers. Thus, it is optimal to use a sampler that has a means of weight adjustment. If a sampler cannot consistently achieve the desired penetration depth, an alternate device should be used.

Once the sampler is secured on board the survey vessel, it is essential that the surface of the sample be made accessible without disturbing the sample. Generally, samplers have hinged flaps on their upper face for this purpose. The opening(s) in the upper face of the sampler should be large enough to allow easy subsampling of the sediment surface. If an opening is too small, the sample may be disturbed as the scientific crew member struggles to take a subsample.

PENETRATION DEPTH

For characterizing surficial sediments in Puget Sound, it is recommended that the upper 2 cm of the sediment column be evaluated. When collecting the upper 2 cm of sediment, it is recommended that a minimum penetration depth of 4-5 cm be achieved for each acceptable sample.

Although the 2-cm specification is arbitrary, it will ensure that:

- Relatively recent sediments are sampled
- Adequate volumes of sediments can be obtained readily for laboratory analyses
- Data from different studies can be compared validly.

Sampling depths other than 2 cm may be appropriate for specific purposes. For example, the upper 1 cm of sediment may be required to determine the age of the most recently deposited sediments. By contrast, a sample depth much greater than 2 cm may be required to evaluate the vertical profile of sediment characteristics or to determine depth-averaged characteristics prior to dredging. If a sampling depth other than 2 cm is used, comparisons with data from 2-cm deep samples may be questionable.

OPERATION OF SAMPLER

The sampling device should be attached to the hydrowire using a ball-bearing swivel. The swivel will minimize the twisting forces on the sampler during deployment and ensure that proper contact is made with the bottom. For safety, the hydrowire, swivel, and all shackles should have a load capacity at least three times greater than the weight of a full sampler.

The sampler should be lowered through the water column at a controlled speed of approximately 1 ft/sec. Under no circumstances should the sampler be allowed to "free fall" to the bottom, as this may result in premature triggering, an excessive bow wake, or improper

orientation upon contact with the bottom. The sampler should contact the bottom gently and only its weight or piston mechanism should be used to force it into the sediment.

After the sediment sample is taken, the sampler should be raised slowly off the bottom and then retrieved at a controlled speed of approximately 1 ft/sec. Before the sampler breaks the water surface, the survey vessel should head into the waves (if present) to minimize vessel rolling. This maneuver will minimize swinging of the sampler after it breaks the water surface. If excessive swinging occurs or if the sampler strikes the vessel during retrieval, extra attention should be paid to evaluating sample disturbance when judging sample acceptability.

The sampler should be secured immediately after it is brought on board the survey vessel. If the sampler tips or slides around before being secured, extra attention should be paid to evaluating sample disturbance.

SAMPLE ACCEPTABILITY CRITERIA

After the sampler is secured on deck, the sediment sample should be inspected carefully before being accepted. The following acceptability criteria should be satisfied:

- The sampler is not over-filled with sample so that the sediment surface is pressed against the top of the sampler
- Overlying water is present (indicates minimal leakage)
- The overlying water is not excessively turbid (indicates minimal sample disturbance)
- The sediment surface is relatively flat (indicates minimal disturbance or winnowing)
- The desired penetration depth is achieved (i.e., 4-5 cm for a 2-cm deep surficial sample).

If a sample does not meet all criteria, it should be rejected.

SAMPLE COLLECTION

After a sample is judged acceptable, the following observations should be noted on the field log sheet:

- Station location
- Depth
- Gross characteristics of the surficial sediment
  - — Texture
  - — Color
  - — Biological structures (e.g., shells, tubes, macrophytes)
  - — Presence of debris (e.g., wood chips, wood fibers, human artifacts)
  - — Presence of oily sheen
  - — Odor (e.g., hydrogen sulfide, oil, creosote)
- Gross characteristics of the vertical profile
  - — Changes in sediment characteristics
- Presence and depth of redox potential discontinuity (rpd) layer
- Penetration depth
- Comments related to sample quality
  - — Leakage
  - — Winnowing
  - — Disturbance.

Before subsamples of the surficial sediments are taken, the overlying water must be removed. The preferred method of removing this water is by slowly siphoning it off near one side of the sampler. Methods such as decanting the water or slightly cracking the grab to let the water run out are not recommended, as they may result in unacceptable disturbance or loss of fine-grained surficial sediment and organic matter.

Once the overlying water has been removed, the surficial sediment can be subsampled. It is recommended that subsamples be taken using a flat scoop shaped like a coal shovel. The shoulders of the scoop should be 2 cm high. This device will allow a relatively large subsample to be taken accurately to a depth of 2 cm. Coring devices are not recommended because generally they collect small amounts of surficial sediment and therefore require repeated extractions to obtain a sufficient volume of material for analysis of conventional sediment variables. A curved scoop is not recommended because it does not sample a uniform depth. Because accurate and consistent subsampling requires practice, it is advisable that an experienced person perform this task.

When subsampling surficial sediments, unrepresentative material should be removed in the field under the supervision of the chief scientist and noted on the field log sheet. The criteria used to determine representativeness should be determined prior to sampling.

Finally, if samples are to be analyzed for trace metals or priority pollutant organic compounds, sample contamination during collection must be avoided. All sampling equipment (i.e., siphon hoses, scoops, containers) should be made of noncontaminating material and should be cleaned appropriately before use. Samples should not be touched with ungloved fingers. In addition, potential airborne contamination (e.g., stack gases, cigarette smoke) should be avoided. Detailed guidance for preventing sample contamination is given in the protocols for metals and organic compounds in other chapters of this notebook.

PARTICLE SIZE

USE AND LIMITATIONS

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. Particle size is also an important variable for marine engineering purposes. In addition to Plumb (1981), a variety of other references discuss the uses and measurement of particle size (e.g., Krumbein and Pettijohn 1938; Folk 1968; Buchanan 1984).

Particle size can be characterized in a wide range of detail. The grossest divisions that generally are considered useful for characterizing particle size distributions are percentages of gravel, sand, silt, and clay. However, each of these size fractions can be subdivided further so that additional characteristics of the size distribution (e.g., mean diameter, skewness, kurtosis) can be determined.

Particle size determinations can either include or exclude organic material. If organic material is removed prior to analysis, the "true" (i.e., primarily inorganic) particle size distribution is determined. If organic material is included in the analysis, the "apparent" (i.e., organic plus inorganic) particle size distribution is determined. Because true and apparent distributions may differ, detailed comparisons between samples analyzed by these different methods are questionable. It is therefore desirable that all samples within each study (at a minimum) and among different studies (if possible) be analyzed using only one of these two methods.

FIELD PROCEDURES

Collection

Samples can be collected in glass or plastic containers. A minimum sample size of 100-150 g is recommended. If unrepresentative material is to be removed from the sample, it should be removed in the field under the supervision of the chief scientist and noted on the field log sheet.

Processing

Samples should be stored at 4° C, and can be held for up to 6 mo before analysis. Samples must not be frozen or dried prior to analysis, as either process may change the particle size distribution.

LABORATORY PROCEDURES

Analytical Procedures

- Equipment
  - Sieve shaker
    - Ro-Tap or equivalent
  - Drying oven
  - Constant temperature bath
  - Analytical balance
    - 0.1 mg accuracy
  - Desiccator
  - Clock
    - With second hand
  - Standard sieves
    - Appropriate mesh sizes
  - Sieve pan and top
  - Sieve brush
  - Funnel
  - 1-L graduated cylinders
  - 50-mL beakers
  - 20-mL pipets
  - Water pique or squirt bottle
  - Glossy paper
  - Dispersant
    - 1 percent sodium hexametaphosphate = 1 percent commercially available Calgon
  - Distilled water.

- Sample preparation
  - Allow samples to warm to room temperature.
  - Homogenize each sample mechanically.
  - Remove a representative aliquot (approximately 25 g) and analyze for total solids content. This information can be used to estimate the dry weight of the aliquot used for particle size analysis. The efficiency of the entire analysis can then be evaluated by adding the dry weights of all sample fractions and comparing this sum with the estimated dry weight of the original aliquot.
  - Remove a second representative aliquot for wet sieving. The aliquot can range from

20 g for muddy sediments to 100 g for sandy sediments. The critical factor for sample size determination is the weight of fine-grained material that will be used for the pipet analysis. Ideally the total dry weight of fine-grained material in the 1-L graduated cylinder should equal approximately 15 g. However, total weights between 5 and 25 g are considered acceptable. Total weights outside this range are not considered acceptable and it is recommended that aliquot size be modified to bring the amount of fine-grained material into the acceptable range.

— Weigh the wet sample to the nearest 0.01 g.

- Organics oxidation - this step removes organic material from the sample. It is optional and depends upon the objectives of each study.
  - — Place the sediment sample in a large beaker (≥ 2 L).
  - — Add 20 mL of 10 percent hydrogen peroxide solution.
  - — Let the sample stand until frothing stops.
  - — Once frothing stops, add an additional 10 mL of hydrogen peroxide solution.
  - — Continue adding 10-mL portions of hydrogen peroxide solution until no frothing occurs on addition.
  - — Boil the sample to remove any excess hydrogen peroxide.
  - — Be careful that material is not lost from the beaker during frothing and boiling.

- Wet-sieving - this step separates the sample into size fractions greater than 62.5 um (i.e., sand and gravel) and less than 62.5 um (i.e., silt and clay)
  - — Place the 62.5-um (4 phi) sieve in a funnel, with a 1-L graduated cylinder underneath. Moisten the sieve using a light spray of distilled water.
  - — Place the sample in a beaker, add 20-30 mL of distilled water, and stir to suspend fine-grained material.
  - — Pour the sample into the sieve and thoroughly rinse the beaker and stirrer with distilled water.
  - — Wash the sediment on the sieve with distilled water using a water pique or squirt bottle having low water pressure. Aggregates can be gently broken using a rubber policeman.
  - — Continue wet sieving until only clear water passes through the sieve. Try to ensure that the rinsate does not exceed approximately 950 mL. This can generally be accomplished by sieving a sample quantity that is not too large and by efficient use of the rinse water. Both of these techniques may require experimentation before routine wet sieving is started.

- Gravel-sand fraction - this fraction is subdivided further by mechanically dry sieving it through a graded series of screens.
  - — Wash the coarse fraction into a preweighed 50-mL beaker using distilled water. Rinse the sieve thoroughly.
  - — Dry the coarse fraction to constant weight at 90 ± 2° C. The drying temperature is less than 100° C to prevent boiling and potential loss of sample.

- Cool the sample to room temperature in a desiccator.
- Weigh the cooled sample to the nearest 0.1 mg.
- Set up a nest of sieves that will divide the coarse fraction into the desired number of subfractions. Set up the sieves in a graded series of mesh sizes, with the coarsest mesh on top and the finest mesh on the bottom. The bottom sieve always should have a mesh size of 62.5 um (4 phi). Place a solid pan on the bottom of the stack and a lid on top of the stack. At a minimum, the coarse fraction should be separated into gravel and sand fractions, using a sieve with a mesh size of 2 mm (-1 phi).
- Add the sample to the uppermost sieve. Complete transfer can be ensured by using a sieve brush to remove any material adhering to the beaker. The sieve brush can also be used to gently break up aggregated sediment.
- Shake mechanically for exactly 15 min using the Ro-Tap (or equivalent). A shaker having an automatic timer is preferable.
- After shaking, empty the contents of each sieve onto a glossy piece of paper (e.g., wax paper). To empty a sieve, invert it and tap it on the table several times while ensuring that all edges hit the table at the same time. If the sieve is not tapped evenly, the meshes may be distorted. After tapping the sieve, ensure complete removal of the sample by brushing the back of the screen. After brushing the back of the screen, turn the sieve over and brush out any particles adhering to the sides of the sieve or the inside of the screen.
- Add the fraction that passed through the bottom sieve (e.g., 4 phi) and was retained by the solid pan to the silt-clay fraction of that sample.
- Weigh each remaining size fraction to the nearest 0.1 mg.
- Sum the weights of all size fractions and compare the result with the initial weight of the coarse fraction. Losses and inaccuracies should be less than 1 percent of the initial weight.
- Large amounts of organically derived fragments (e.g., wood debris, grass, shells) or any unusual material in any size fraction should be noted on the laboratory log sheet.

• Silt-clay fraction - this fraction is subdivided further using a pipet technique that depends upon the differential settling rates of different particles. Because additions to this fraction may be made after mechanical sieving of the gravel-sand fraction (see above), it is recommended that the silt-clay analysis for each sample not be conducted until the gravel-sand analysis has been completed.

- Add 10 mL of the dispersant to 990mL of distilled water. Determine the weight of dispersant in a 20-mL aliquot by pipeting a 20-mL aliquot of dispersant into each of five tared beakers, drying the samples to constant weight at 90 ± 2° C, cooling the samples in a desiccator, weighing the cooled samples, and calculating the mean weight of dispersant in the five samples. This weight will be subtracted from the weight of each sediment fraction at the end of the pipet analysis.
- Add 10 mL of the dispersant to the each sample suspension in the 1-L graduated cylinders.

- Mix each suspension by either stoppering and inverting the cylinder or by using the up and down motion of a perforated disc plunger.
- Allow the mixed suspension to stand for 2-3 h and check for signs of flocculation. Flocculation can be recognized by a curdling and rapid settling of lumps of particles or by the presence of a thick soupy layer on the bottom of the cylinder passing abruptly into clear water above.
- If flocculation occurs, add dispersant in 10-mL increments until no noticeable flocculation occurs. Record the volume of dispersant added.
- When ready to conduct the pipet analysis, bring the sample volume to 1 L by adding distilled water, mix the suspension thoroughly, and place the cylinder in a constant-temperature water bath. If the volume is greater or less than 1 L, the factor for converting the weight of the sediment in each 20-mL aliquot to that in the total volume must be modified accordingly.
- After 20 sec, withdraw a 20-mL aliquot from a depth of 20 cm below the surface of the suspension using a pipet. The pipet should be marked for the specified sampling depths and should be inserted vertically into the settling cylinder when the aliquot is taken. A suction bulb may be used on the open end of the pipet to facilitate sampling. It is critical that the suspension be disturbed as little as possible when pipet aliquots are taken.
- Transfer the 20-mL aliquot to a preweighed 50-mL beaker. Rinse the pipet into the beaker using 20 mL of distilled water.
- Withdraw 20-mL aliquots at a depth of 10 cm below the surface of the suspension at the appropriate time(s) listed in Table 3. A formula for calculating withdrawal times is given by Folk (1968) and Buchanan (1984). If a withdrawal is missed, the suspension can be stirred again and the missed withdrawal can be taken at the appropriate time after settling begins. It is not necessary to withdraw the initial 20-mL aliquot when this corrective action is conducted.
- Transfer these additional 20-mL aliquots to 50-mL preweighed beakers, each time rinsing the pipet into the respective beaker using 20 mL of distilled water
- Dry all aliquots to constant weight at 90 ± 2° C. A drying temperature less than 100° C is used to prevent boiling and potential loss of sample.

TABLE 3. Withdrawal times for pipet analysis as a function of particle size and water temperature[ab]

Diameter finer than		Withdrawal depth	Elapsed time for Withdrawal of Sample in Hours (h), Minutes (m) and Seconds (s)						
(phi)	(µ)	(cm)	18°C	19°C	20°C	21°C	22°C	23°C	24°C
4.0	62.5	20	20s	20s	20s	20s	20s	20s	20s
5.0	31.2	10	2m 0s	1m 57s	1m 54s	1m 51s	1m 49s	1m 46s	1m 44s
6.0	15.6	10	8m 0s	7m 48s	7m 36s	7m 25s	7m 15s	7m 5s	6m 55s
7.0	7.8	10	31m 59s	31m 11s	30m 26s	29m 41s	28m 59s	28m 18s	27m 39s
8.0[c]	3.9	10	2h 8m	2h 5m	2h 2m	1h 59m	1h 56m	1h 53m	1h 51m
9.0	1.95	10	8h 32m	8h 18m	8h 6m	7h 56m	7h 44m	7h 32m	7h 22m
10.0	0.98	10	34h 6m	33h 16m	32h 28m	31h 40m	30h 56m	30h 12m	29h 30m

[a] Modified from Plumb (1981)
[b] It is critical that temperature be held constant during the pipet analysis.
[c] Breakpoint between silt and clay.

— Cool dried samples to room temperature in a desiccator.
— Weigh cooled samples to the nearest 0.1 mg.

- Calculations
  — The total weight of a phi-size interval in the 1-L graduated cylinder is determined as follows:

$$\Phi\text{weight (g dry wt)} = 50\,[(A - C) - (B - C)]$$

Where:
A = weight (g) of residue in a 20-mL aliquot for a given phi-size boundary
B = weight (g) residue in a 20-mL aliquot for the next larger phi-size boundary
C = mean weight (g) of dispersant in a 20-mL aliquot.

QA/QC Procedures

It is critical that each sample be homogenized thoroughly in the laboratory before a subsample is taken for analysis. Laboratory homogenization should be conducted even if samples were homogenized in the field.

After dry-sieving a sample, all material must be removed from the sieve. This can be accomplished by tapping the rim of the sieve evenly on a hard surface and by brushing the screen.

The total amount of fine-grained material used for pipet analysis should be 5-25 g. If more material is used, particles may interfere with each other during settling and the possibility of flocculation may be enhanced. If less material is used, the experimental error in weighing becomes unacceptably large.

Before pipet extractions can be made, the sample must be homogenized thoroughly within the settling cylinder. Once the pipet analysis begins, the settling cylinders must not be disturbed, as this will alter particle settling velocities. Care must be taken to disturb the sample as little as possible when pipet extractions are made.

After a pipet extract has been transferred to a drying beaker, any sample adhering to the inside of the pipet must be removed. This can be accomplished by drawing 20 mL of distilled water into the pipet and adding this rinse water to the drying beaker.

Dried samples should be cooled in a desiccator and held there until they are weighed. If a desiccator is not used, the sediment will accumulate ambient moisture and the sample weight will be overestimated. A color-indicating desiccant is recommended so that spent desiccant can be detected easily. Also, the seal on the desiccator should be checked periodically, and, if necessary, the ground glass rims should be greased or the "O" rings should be replaced.

It is recommended that triplicate analyses be conducted on one of every 20 samples, or on one sample per batch if less than 20 samples are analyzed. It is also recommended that the analytical balance, drying oven, and temperature bath be inspected daily and calibrated at least once per week.

DATA REPORTING REQUIREMENTS

The weight of each sediment fraction should be reported to the nearest 0.0001 g dry weight. The laboratory should report the results of all samples analyzed (including QA replicates) and should note any problems that may have influenced data quality.

TOTAL SOLIDS

USE AND LIMITATIONS

Total solids are the organic and inorganic materials remaining after a sample has been dried completely. This variable is commonly used to convert sediment concentrations of substances from a wet-weight to a dry-weight basis.

Total solids values are operationally defined, because results depend on drying temperatures. For example, temperature-dependent weight losses occur from volatilization of organic matter, mechanically occluded water, water of crystallization, and gases from heat-induced chemical decomposition. By contrast, weight gains may result from oxidation processes. To provide data that are comparable among different studies, it is therefore critical that drying temperatures be standardized.

FIELD PROCEDURES

Collection

Samples can be collected in glass or plastic containers. A minimum sample size of 50 g is recommended. If unrepresentative material is to be removed from the sample, it should be removed in the field under the supervision of the chief scientist and noted on the field log sheet.

Processing

Samples should be stored frozen and can be held for up to 6 mo under that condition.

LABORATORY PROCEDURES

Analytical Procedures

- Equipment
  - Muffle furnace
    - 550° C capacity
  - Drying oven
  - Desiccator
  - Analytical balance
    - 0.01 g accuracy
  - 100-mL evaporating dishes
    - Porcelain, platinum, or Vycor.

- Equipment preparation

- Ignite clean evaporating dishes at 550 ± 10° C for 1 h in a muffle furnace to remove any remaining organic material.
- Cool ignited dishes to room temperature in a desiccator.
- Weigh each cooled dish to the nearest 0.01 g and store in the desiccator.

• Sample preparation
- Allow frozen sediment samples to warm to room temperature
- Homogenize each sample mechanically.
- Transfer a representative subsample (approximately 25 g) to a preweighed evaporation dish.
- Weigh the undried sample to the nearest 0.01 g.

• Analytical procedures
- Dry the sample to constant weight at 103 ± 2° C.
- Cool the dried sample to room temperature in a desiccator.
- Weigh the cooled sample to the nearest 0.01 g.

• Calculations
- Total solids content is determined as follows:

$$\text{Percent solids} = \frac{(A-B)\,(100)}{C-B}$$

Where:
A = weight (g) of dish and dry sample residue
B = weight (g) of dish
C = weight (g) of dish and wet sample.

QA/QC Procedures

It is critical that each sample be thoroughly homogenized in the laboratory before a subsample is taken for analysis. Laboratory homogenization should be conducted even if samples were homogenized in the field.

Evaporating dishes must be ignited at 550° C before being used for total solids analysis. This step ensures that dishes are free from organic contaminants.

Dried samples should be cooled in a desiccator and held there until they are weighed. If a desiccator is not used, the sediment will accumulate ambient moisture and the sample weight will be overestimated. A color-indicating desiccant is recommended so that spent desiccant can be detected easily. Also, the seal on the desiccator should be checked periodically and, if necessary, the ground glass rims should be greased or the "O" rings should be replaced.

It is recommended that triplicate analyses be conducted on one of every 20 samples or on one sample per batch if less than 20 samples are analyzed. It is also recommended that the analytical balance and drying oven be inspected daily and calibrated at least once per week.

DATA REPORTING REQUIREMENTS

Total solids should be reported as a percentage of the wet weight of the sample to the nearest 0.1 unit. The laboratory should report the results of all samples analyzed (including QA replicates) and should note any problems that may have influenced sample quality.

TOTAL VOLATILE SOLIDS (TVS)

USE AND LIMITATIONS

Total volatile solids represent the fraction of total solids that are lost on ignition at a higher temperature than that used to determine total solids. Total volatile solids is used as a crude estimate of the amount of organic matter in the total solids.

Total volatile solids is operationally defined by the ignition temperature. Total volatile solids content does not always represent the organic content of a sample because some organic material may be lost at the drying temperature and some inorganic material (e.g., carbonates, chlorides) may be lost at the ignition temperature. Because of the temperature dependence of total volatile solids, valid interstudy comparisons require the use of standardized drying and ignition temperatures.

FIELD PROCEDURES

Collection

Samples can be collected in glass or plastic containers. A minimum sample size of 50 g is recommended. If unrepresentative material is to be removed from the sample, it should be removed in the field under the supervision of the chief scientist and noted on the field log sheet.

Processing

Samples should be stored frozen and can be held for up to 6 mo under that condition.

LABORATORY PROCEDURES

Analytical Procedures

- Equipment
  - Muffle furnace
    - 550° C capacity
  - Drying oven
  - Desiccator
  - Analytical balance
    - 0.01 g accuracy
  - 100-mL evaporating dishes
    - Porcelain, platinum, or Vycor.

- Equipment preparation
  - Ignite clean evaporating dishes at 550 ± 10° C for 1 h in a muffle furnace to remove any remaining organic material.
  - Cool ignited dishes to room temperature in a desiccator.
  - Weigh each cooled dish to the nearest 0.01 g and store in the desiccator.

- Sample preparation
  - Allow frozen sediment samples to warm to room temperature.
  - Homogenize each sample mechanically.
  - Transfer a representative subsample (approximately 25 g) to a preweighed evaporating dish.

- Analytical procedures
  - Dry the sample to constant weight at 103 ± 2° C.
  - Cool the dried sample to room temperature in a desiccator.
  - Weigh the cooled sample to the nearest 0.01 g.
  - Ignite the sample at 550 ± 10° C to constant weight. Make sure that the samples do not flare up when placed in the oven, as sediment may be lost from the crucibles. If sample flashing is a problem, it is recommended that the muffle furnace be cooler than 550° C when samples are placed inside, and that the temperature gradually be increased to 550° C.
  - Weigh each cooled sample to the nearest 0.01 g.

- Calculations
  - TVS content is determined as follows:

$$\text{Percent TVS} = \frac{(A-C)100}{A-B}$$

Where:

A = weight (g) of dish and dry sample residue
B = weight (g) of evaporation dish
C = weight (g) of dish and ignition residue.

QA/QC Procedures

It is critical that each sample be thoroughly homogenized in the laboratory before a subsample is taken for analysis. Laboratory homogenization should be conducted even if samples were homogenized in the field.

Evaporating dishes (or crucibles) must be ignited at 550° C before being used for total volatile solids analysis. This step ensures that the dishes are free from volatile contaminants.

Dried and combusted samples should be cooled in a desiccator and held there until they are weighed. If a desiccator is not used, the sediment will accumulate ambient moisture and the sample weight will be overestimated. A color-indicating desiccant is recommended so that spent desiccant can be detected easily. Also, the seal on the desiccator should be checked periodically and, if necessary, the ground glass rims should be greased or the "O" rings should be replaced.

It is recommended that triplicate analyses be conducted on one of every 20 samples or on one sample per batch if less than 20 samples are analyzed. It is also recommended that the analytical balance, drying oven, and muffle furnace be inspected daily and calibrated at least once per week.

DATA REPORTING REQUIREMENTS

Total volatile solids should be reported as a percentage of the dry weight of the uncombusted sample to the nearest 0.1 unit. The laboratory should report the results of all samples analyzed (including QA replicates) and should note any problems that may have influenced data quality.

TOTAL ORGANIC CARBON (TOC)

USE AND LIMITATIONS

Total organic carbon is a measure of the total amount of nonvolatile, volatile, partially volatile, and particulate organic compounds in a sample. Total organic carbon 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 biochemical and chemical oxygen demand tests.

Because inorganic carbon (e.g., carbonates, bicarbonates, free CO_2) will interfere with total organic carbon determinations, samples should be treated to remove inorganic carbon before being analyzed.

FIELD PROCEDURES

Collection

Samples can be collected in glass or plastic containers. A minimum sample size of 25 g is recommended. If unrepresentative material is to be removed from the sample, it should be removed in the field under the supervision of the chief scientist and noted on the field log sheet.

Processing

Samples should be stored frozen and can be held for up to 6 mo under that condition. Excessive temperatures should not be used to thaw samples.

LABORATORY PROCEDURES

Analytical Procedures

- Equipment
  - Induction furnace
    - e.g., Leco WR-12, Dohrmann DC-50, Coleman CH analyzer, Perkin Elmer 240 elemental analyzer, Carlo-Erba 1106
  - Analytical balance
    - 0.1 mg accuracy
  - Desiccator
  - Combustion boats
  - 10 percent hydrochloric acid (HCl)
  - Cupric oxide fines
  - Benzoic acid.

- Equipment preparation
  - Clean combustion boats by placing them in the induction furnace at 950° C. After being cleaned, combustion boats should not be touched with bare hands.
  - Cool boats to room temperature in a desiccator.
  - Weigh each boat to the nearest 0.1 mg.

- Sample preparation
  - Allow frozen samples to warm to room temperature.
  - Homogenize each sample mechanically.
  - Transfer a representative aliquot (5-10 g) to a clean container.

- Analytical procedures
  - Dry samples to constant weight at 70 ± 2° C. The drying temperature is relatively low to minimize loss of volatile organic compounds.
  - Cool dried samples to room temperature in a desiccator.
  - Grind sample using a mortar and pestle to break up aggregates.
  - Transfer a representative aliquot (0.2-0.5 g) to a clean, preweighed combustion boat.
  - Determine sample weight to the nearest 0.1 mg.
  - Add several drops of HCl to the dried sample to remove carbonates. Wait until the effervescing is completed and add more acid. Continue this process until the incremental addition of acid causes no further effervescence. Do not add too much acid at one time as this may cause loss of sample due to frothing. Exposure of small samples (i.e., 1-10 mg) having less than 50 percent carbonate to an HCl atmosphere for 24-48 h has been shown to be an effective means of removing carbonates (Hedges and Stern 1984). If this method is used for sample sizes greater than 10 mg, its effectiveness should be demonstrated by the user.
  - Dry the HCl-treated sample to constant weight at 70 ± 2° C.
  - Cool to room temperature in a desiccator.
  - Add previously ashed cupric oxide fines or equivalent material (e.g., alumina oxide) to the sample in the combustion boat.
  - Combust the sample in an induction furnace at a minimum temperature of 950 ± 10° C.

- Calculations
  - If an ascarite-filled tube is used to capture CO_2, the carbon content of the sample can be calculated as follows:

$$\text{Percent Carbon} = \frac{A(0.2729)(100)}{B}$$

Where:

A = the weight (g) of CO_2 determined by weighing the ascarite tube before and after combustion
B = dry weight (g) of the unacidified sample in the combustion boat

0.2729 = the ratio of the molecular weight of carbon to the molecular weight of carbon dioxide

A silica gel trap should be placed before the ascarite tube to catch any moisture driven off during sample combustion. Additional silica gel should be placed at the exit end of the ascarite tube to trap any water that might be formed by reaction of the trapped CO_2 with the NaOH in the ascarite.

— If an elemental analyzer is used, the amount of CO_2 will be measured by a thermal conductivity detector. The instrument should be calibrated daily using an empty boat blank as the zero point and at least two standards. Standards should bracket the expected range of carbon concentrations in the samples.

QA/QC Procedures

It is critical that each sample be thoroughly homogenized in the laboratory before a subsample is taken for analysis. Laboratory homogenization should be conducted even if samples were homogenized in the field.

Dried samples should be cooled in a desiccator and held there until they are weighed. If a desiccator is not used, the sediment will accumulate ambient moisture and the sample weight will be overestimated. A color-indicating desiccant is recommended so that spent desiccant can be detected easily. Also, the seal on the desiccator should be checked periodically and, if necessary, the ground glass rims should be greased or the "O" rings should be replaced.

It is recommended that triplicate analyses be conducted on one of every 20 samples, or on one sample per batch if less than 20 samples are analyzed. A method blank should be analyzed at the same frequency as the triplicate analyses. The analytical balance should be inspected daily and calibrated at least once per week. The carbon analyzer should be calibrated daily with freshly prepared standards. A standard reference material should be analyzed at least once for each major survey.

DATA REPORTING REQUIREMENTS

Total organic carbon should be reported as a percentage of the dry weight of the unacidified sample to the nearest 0.1 unit. The laboratory should report the results of all samples (including QA replicates, method blanks, and standard reference measurements) and should note any problems that may have influenced sample quality. The laboratory should also provide a summary of the calibration procedure and results (e.g., range covered, regression equation, coefficient of determination).

OIL AND GREASE (FREON-EXTRACTABLE)

USE AND LIMITATIONS

Oil and grease tests measure all material recovered as a substance soluble in a nonpolar solvent (e.g., Freon) under acidic conditions. Oil and grease includes such compounds as hydrocarbons, vegetable oils, animal fats, waxes, soaps, greases, and related industrial compounds.

In addition to oil and grease, the solvent may dissolve other kinds of substances, such as sulfur compounds, organic dyes, and chlorophyll. Oil and grease is therefore operationally defined by the kind of solvent and analytical methods used. Standardized procedures are essential for valid interstudy comparisons.

FIELD PROCEDURES

Collection

Samples should be collected only in glass containers having TFE-lined lids. Although aluminum-lined lids can be used, seawater eventually will corrode the aluminum. Before being used, containers and lids should first be washed with a warm aqueous detergent mixture and then, in sequence, thoroughly rinsed with hot tap water, rinsed at least twice with distilled water, rinsed once with Freon (i.e., 1,1,2-trichloro-1,2,2-trifluoroethane, and dried at 105 ± 2° C for 30 min. A minimum sample size of 100 g is recommended. If unrepresentative material is to be removed from the sample, it should be removed in the field under the supervision of the chief scientist and noted on the field log sheet.

Processing

If samples cannot be analyzed within 24 h, they can be preserved with approximately 1 mL of concentrated hydrochloric acid (HCl) per 80 g of sample. Acid-preserved samples should be stored at 4° C, and can be held for up to 28 days in that condition. Although U.S. EPA has not established a recommended maximum holding time for oil and grease sediments, 28 days is consistent with the recommended holding time for acid-preserved water samples. Samples can also be preserved by freezing at -20□C, and can be held under that condition for up to 6 mo. Samples must be kept field-moist during storage because they may lose apparent oil and grease as a result of drying.

LABORATORY PROCEDURES

Analytical Procedures

- Equipment
  - Infrared spectrophotometer

IR technique only

— Analytical balance
  Gravimetric technique only, 0.1 mg accuracy
— Extraction apparatus, Soxhlet
— Vacuum pump or other source of vacuum
— Extraction thimble, paper
— Concentrated hydrochloric acid (HCl) or concentrated sulfuric acid (H_2SO_4)
— Magnesium sulfate monohydrate
  Prepare $MgSO_4 \cdot H_2O$ by drying a thin layer of $MgSO_4 \cdot 7H_2O$ overnight in an oven at 103° C
— Freon (1,1,2-trichloro-1,2,2,-trifluoroethane), boiling point 47° C
  The solvent should leave no measurable residue on evaporation; redistill if necessary
— Grease-free cotton
  Extract nonabsorbent cotton using Freon
— Oil reference standard
  If the identity of oil and grease in a sample is unknown, a mixture of 15.0 mL n-hexadecane, 15.0 mL isooctane, and 10.0 mL chlorobenzene should be used as the standard. This is the same reference oil used for water samples in U.S. EPA Method 413.2 (U.S. EPA 1983). If the identity of oil and grease is known, the standard can be comprised of the same substance as that in the sample.

- Sample preparation
  — Allow samples to warm to room temperature.
  — Homogenize each sample mechanically.
  — Remove a representative aliquot (approximately 25 g) and analyze it for total solids content.
  — Remove a representative aliquot (approximately 20 g) and weigh it to the nearest 0.1 mg.
  — Transfer the weighed aliquot to a 150-mL beaker for oil and grease analysis.

- Oil and grease extraction
  — Acidify the sample to pH = 2 using concentrated HCl or concentrated H_2SO_4.
  — Add 25 g $MgSO_4 \cdot H_2O$ to the acidified sediment sample. Stir to make a uniformly smooth paste that is spread on the beaker wall. Allow to stand 15-30 min until solidified.
  — Following solidification, remove the solids and grind in a porcelain mortar. The use of a desiccated, uniformly ground sample improves the efficiency of the extraction process.
  — Add the ground sample to a paper extraction thimble. The beaker and mortar should be wiped with a small piece of filter paper that has been soaked in Freon. Add the filter paper to the paper thimble.
  — Fill the thimble with glass wool or small glass beads. Extract the prepared sample using Freon in a Soxhlet apparatus at a rate of 20 cycles/h for 4 h. If the final extract is turbid, filter the sample through grease-free cotton into a clean flask. Rinse the initial sample container and the cotton with Freon and add the washing to the filtered sample.

— Oil and grease concentration of the extract can be determined using either the infrared spectrophotometry or the gravimetric method.

- Infrared spectrophotometry method
  - — Quantitatively transfer the sediment extract to a convenient size volumetric flask and dilute to volume with Freon.
  - — Prepare calibration standards using the reference oil.
  - — Transfer required amounts of the reference material into 100-mL volumetric flasks using microliter pipettes. Dilute to volume with Freon.
  - — The most appropriate pathlength for the quartz cells to be used in the spectrophotometric determination is determined by the expected sample concentration. The following information is presented as a guide for selecting cell length:

Pathlength, cm		Expected Range, mg
1		4 - 40
5		0.5 - 8
10		0.1 - 4

Based on observed ranges of oil and grease in sediments, it may be necessary to dilute the sample extracts to the working ranges indicated above.

— Scan the standards and samples from 3,200 to 2,700 cm^{-1} using a recording infrared spectrophotometer. Freon should be used in the reference beam of a dual beam instrument or to zero a single beam instrument. The absorbance of the 2930-cm^{-1} peak should be used to construct a standard curve.

— Prepare a standard curve by plotting measured absorbance vs. oil and grease concentration of the standards. Compare the absorbance of the Freon extract to the standard curve to determine the oil and grease concentration.

— Calculate oil and grease concentration as follows:

$$\text{Concentration (mg/kg dry weight)} = \frac{(X)\,(V)\,(1{,}000)}{(g)\,(\%\,S)}$$

Where:

X = concentration of oil and grease in the Freon extract, mg/L
V = volume of Freon extract, L
g = wet weight of sediment extracted, g
%S = percent total solids in the sediment sample expressed as a decimal fraction).

- Gravimetric method
  - Quantitatively transfer the sediment extract to a tared distilling flask. Rinse the extract container with Freon and add to the distilling flask.
  - Distill the Freon from the extraction flasks using a water bath at 70° C.
  - After the solvent has been evaporated, place the flask on a warm steam bath for 15 min and draw air through the flask by means of an applied vacuum for the final 1 min.
  - Cool the sample to constant weight in a desiccator.
  - Weigh the cooled sample to the nearest 0.1 mg.
  - Calculate oil and grease concentration as follows:

$$\text{Concentration (mg/kg dry weight)} = \frac{(A-B)\,(1000)}{(g)\,(\%S)}$$

Where:

A = weight (mg) of tared flask and oil and grease residue
B = weight (mg) of tared flask
C = calculated residue (mg) based on Freon flask
g = wet weight (g) of sediment extracted
%S = percent total solids in the sediment sample (expressed as a decimal fraction).

QA/QC Procedures

Because the results of an oil and grease analysis are extremely sensitive to the methods used, comparable results can be obtained only by strict adherence to all methodological details.

It is critical that each sample be thoroughly homogenized in the laboratory before a subsample is taken for analysis. Laboratory homogenization should be conducted even if samples were homogenized in the field.

It is recommended that triplicate analyses be conducted on one of every 20 samples, or on one sample per batch if less than 20 samples are analyzed. Also, a method blank should be analyzed at the same frequency as the triplicate analyses.

DATA REPORTING REQUIREMENTS

Oil and grease concentrations should be reported as mg/kg dry weight to no more than three significant figures. The laboratory should report the results of all samples (including QA replicates and method blanks) and should note any problems that may have influenced sample quality. The laboratory should also provide a description of the calibration procedures and standards used to determine oil and grease concentrations by infrared spectrophotometry.

TOTAL SULFIDES

USE AND LIMITATIONS

Total sulfides represent the amount of acid-soluble H_2S, HS^-, and S^{2-} in a sample. Sulfides are measured because they may be toxic and because they may create unaesthetic conditions. This method cannot be used if a measure of only water-soluble sulfides is desired. A measure of water-soluble sulfides might be desired if an estimate of biologically available sulfides is needed.

Sulfides are difficult to sample because some may be lost through volatilization and/or gas stripping and some may be lost through oxidation by dissolved oxygen.

FIELD PROCEDURES

Collection

Samples can be collected in glass or plastic containers. A minimum sample size of 50 g is recommended. If unrepresentative material is to be removed from the sample, it should be removed in the field under the supervision of the chief scientist and noted on the field log sheet.

Processing

Samples should be stored at 4° C immediately after collection and analyzed as soon as possible. Samples must be kept moist during storage because oxidation may result from drying. Although U.S. EPA has not established a recommended maximum holding time for sulfides in sediments, a maximum holding time of 7 days would be consistent with the holding time recommended for sulfides in preserved water samples.

LABORATORY PROCEDURES

Analytical Procedures

- Equipment
  - Distillation apparatus, all glass
    For large samples, a suitable assembly consists of a 1-L pyrex distilling flask with Graham condenser as used for the analysis of phenols. A section of glass tubing should be connected to the tip of the condenser so that it reaches the bottom of the collection tube.
  - Distillate collection tubes
    Short-form Nessler tubes, graduated at 50 and 100 mL.
  - Spectrophotometer
    For use at 650 nm and providing a light path of 1 in or greater.

— Nitrogen, water-pumped
— Zinc acetate, 2 N
  Dissolve 220 g of $Zn(C_2H_3O_2)_2 \cdot 2H_2O$ in distilled water and dilute to 1 L.
— Zinc acetate, 0.2 N
  To 100 mL of 2 N $Zn(C_2H_3O_2)_2 \cdot 2H_2O$ add several drops of acetic acid and dilute to 1 L.
— Sulfuric acid solution, 1:1
  Carefully add 500 mL of concentrated H_2SO_4 to 500 mL of distilled water in a 1-L flask. Mix continuously and cool under running water while combining reagents. Cool solution before using.
— Dilute sulfuric acid solution, approximately 0.1 N
  Dilute 5 mL of 1:1 H_2SO_4 to 1 L with distilled water.
— Stock amine solution
  Dissolve 2.7 g of N, N-dimethyl-p-phenylenediamine sulfate and dilute to 100 mL with 1:1 H_2SO_4 solution. This solution is stable for approximately 1 wk.
— Working amine solution
  Dilute 2 mL of stock amine solution to 100 mL with 1:1 H_2SO_4 solution. Prepare fresh daily.
— Ferric chloride solution
  Dissolve 100 g of $FeCl_3 \cdot 6H_20$ in hot distilled water and dilute to 100 mL. Cool before use.
— Standard potassium biniodate solution, 0.025 N
  Accurately weigh out 0.8124 g $KH(IO_3)_2$ and dissolve in distilled water. Dilute to 1 L.
— Standard sodium thiosulfate titrant, 0.025 N
  Dissolve 6.205 g $Na_2S_2O_3 \cdot 5H_20$ in distilled water and dilute to 1 L. Preserve with 5 mL chloroform. Standardize against standard potassium biniodate using starch as an indicator.
— Potassium iodide solution
  Dissolve 5 g of KI in distilled water and dilute to 100 mL.
— Treated hydrochloric acid
  Place one or two strips of aluminum in a small beaker of concentrated HCl. Following the subsequent reaction, the acid is poured off and is ready to use.
— Oxygen-free dilution water
  Pass nitrogen gas through a sufficient quantity of distilled water for dilution requirements. A minimum of 10 min is required to displace oxygen in the water.
— Sodium sulfide, reagent, crystal.

- Standards preparation
  — Prepare 0.01 N sulfide solution as follows: weigh approximately 1.2 g of large crystal $Na_2S \cdot 9H_20$. Wash the crystals several times with distilled water. Discard the washings and add the washed crystals to 975 mL of nitrogen-saturated distilled water. Dilute to 1 L.

- Pipet 20 mL of stock sulfide solution into 100 mL of oxygen-free water. Add 5 mL of KI solution, 20 mL of 0.025 N $KH(IO_3)_2$ solution, and 10 mL of 0.1 N H_2SO_4. Titrate with 0.025 N $Na_2S_20_3$ solution using starch as an endpoint indicator. Carry a blank through the procedure and calculate the amount of reacted iodine from the difference between the blank and standard titrations. Because 1 mL of 0.025 N KH $(IO_3)_2$ is equivalent to 0.400 mg of sulfide ion, calculate the sulfide concentration in the stock solution.
- Calculate the volume of stock solution that contains 0.2 mg sulfide and add this amount to 900 mL of oxygen-free water. Dilute to 1 L. This is the working standard containing 2 ug S/mL. Sulfide solutions are extremely unstable and must be prepared fresh and used immediately. Stability is increased by using nitrogen-saturated water for dilution.
- Prepare a standard curve by dilution of the working sulfide solution. Pipet 20 mL 0.2 N $Zn(C_2H_30_2)_2$ into a series of 50-mL Nessler tubes. Add the required amounts of sulfide solution to each Nessler tube, taking care to pipet the solution below the $Zn(C_2H_30_2)_2$ level. Dilute to 50 mL with oxygen-free water.
- Equilibrate the temperature of the standards to 23-25° C using a water bath while the colorimetric reagents are added. Add 2 mL dilute amine-sulfuric acid solution to the standard, mix, and add 0.25 mL (5 drops) $FeCl_3$ solution. Mix the solution and allow 10 min for color development. Measure the absorbance at 650 nm.

- Sample preparation
  - Allow samples to warm to room temperature.
  - Homogenize each sample mechanically.
  - Remove a representative aliquot (approximately 25 g) and analyze for total solids content.
  - Remove a representative aliquot for total sulfides analysis. The aliquot should not contain more than 50 ug of sulfide.
  - Weigh the aliquot to the nearest 0.1 mg.

- Distillation
  - Set up the distillation apparatus. The transfer tube from the condenser should reach the bottom of the distillated collection tube. The condenser should be attached so that it can be easily moved up or down when diluting the distillate or adding reagents.
  - Pipet 20 mL of 0.2 N $Zn(C_2H_30_2)_2$ into a 100-mL Nessler tube and lower the condenser so that the transfer tubing reaches below the level of the liquid. Attach a distilling flask and pass nitrogen gas through the system for at least 10 min.
  - Transfer the sample aliquot to the distillation flask. Bubble nitrogen gas through the sample to remove any oxygen dissolved in the sample. A small amount of sulfide may be driven over by the gas, so be sure that the only exit is through the zinc acetate solution in the collecting tube.
  - Discontinue nitrogen evolution and add rapidly several boiling stones, two drops of methyl orange indicator, and enough treated HCl to change the color from orange to red. Stopper as quickly as possible and heat slowly. The slower the heating rate, the greater

the contact time between the evolved H_2S and $Zn(C_2H_3O_2)_2$ and the less chance of sulfide loss.

— Distill the solution until approximately 20 mL of distillate has been collected (roughly 5-8 min after the solution commences to boil). Turn off heat and remove the stopper in the distillation flask to keep the distillate from being sucked back up the condenser. Raise the transfer tube above the 50-mL mark on the collection container and dilute the solution to 50 mL.

— Place the distillates in a water bath at 23-25° C. Add 2 mL dilute amine solution and mix. Add 0.25 mL (5 drops) $FeCl_3$ solution and mix. Allow 10 min for color development and measure sample absorbance at 650 nm.

- Calculations

— Prepare a standard curve by plotting absorbance of the standards vs. sulfide concentration. Ensure that the standards cover the range of concentrations expected in the samples. Determine the sulfide concentration of the sample distillate by comparing sample absorbance with the standard curve.

— Calculate total sulfides concentration as follows:

$$\text{Concentration (mg/kg dry weight)} = \frac{(C)\ (0.05)\ (1000)}{(g)\ (\%S)}$$

Where:

C = sulfide concentration in distillate, mg/L
0.05 = sample volume of distillate, L (as written)
g = wet weight of sediment aliquot, g
%S = percent solids of sediment as a decimal fraction.

QA/QC Procedures

It is critical that each sample be thoroughly homogenized in the laboratory before a subsample is taken for analysis. Laboratory homogenization should be conducted even if samples were homogenized in the field.

It is recommended that triplicate analyses be conducted on one of every 20 samples, or on one sample per batch if less than 20 samples are analyzed. Fresh standards should be used to calculate a calibration curve for each batch of samples. The analytical balance should be inspected daily and calibrated at least once per week.

DATA REPORTING REQUIREMENTS

Total sulfides should be reported as mg/kg of sediment dry weight to the nearest 0.1 unit. The laboratory should report the results of all samples (including QA replicates) and should note any problems that may have influenced sample quality. The laboratory should also describe the calibration curve used to determine total sulfide concentrations.

TOTAL NITROGEN

If the elemental analyzer used to measure total organic carbon can also measure total nitrogen, it is recommended that the latter variable be measured simultaneously with TOC. Total nitrogen values in sediments generally are used to compare carbon-to-nitrogen ratios. A separate total nitrogen analysis using a technique other than the elemental analyzer method is not considered equivalent for calculating carbon-to-nitrogen ratios.

BIOCHEMICAL OXYGEN DEMAND (BOD)

USE AND LIMITATIONS

Biochemical oxygen demand is a 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 (e.g., total volatile solids, total organic carbon, chemical oxygen demand).

In addition to oxygen used for degrading organic matter, biochemical oxygen demand may also include oxygen used to oxidize inorganic material (e.g., sulfide, ferrous iron) and reduced forms of nitrogen.

FIELD PROCEDURES

Collection

Samples can be collected in glass or plastic containers. A minimum sample size of 50 g is recommended. If unrepresentative material is to be removed from the sample, it should be removed in the field under the supervision of the chief scientist and noted on the field log sheet.

Processing

Samples should be stored at 4° C, and can be held for up to 7 days under that condition. Samples should be kept field-moist and air contact should be prevented to minimize oxidation.

LABORATORY PROCEDURES

Analytical Procedures

- Equipment
  - Incubator
    - Thermostatically controlled at 20° ± 1° C.
    - All light should be excluded to prevent the photosynthetic production of dissolved oxygen by algae in the sample.
  - Incubation bottles
    - 300-mL capacity, with ground glass stoppers.
  - Distilled water
    - Free of copper, chlorine, chloramines, caustic alkalinity, acids, and organic material.
  - Phosphate buffer solution

Dissolve the following in distilled water: 8.5 g potassium dihydrogen phosphate, KH_2PO_4; 21.75 g dipotassium hydrogen phosphate, K_2HPO_4; 33.4 g disodium hydrogen phosphate heptahydrate, $Na_2HPO_4 \cdot 7H_2O$; and 1.7 g ammonium chloride, NH_4Cl. Dilute to 1 L. The pH of this buffer should be 7.2 without further adjustment. If dilution water is to be stored in the incubator, the phosphate buffer should be added just prior to using the dilution water.

— Magnesium sulfate solution

Dissolve 22.5 g $MgSO_4 \cdot 7H_2O$ in distilled water and dilute to 1 L.

— Calcium chloride solution

Dissolve 27.5 g anhydrous $CaCl_2$ in distilled water and dilute to 1 L.

— Ferric chloride solution

Dissolve 0.25 g $FeCl_3 \cdot 6H_2O$ in distilled water and dilute to 1 L.

— Dilution water

Store distilled water in cotton-plugged bottles for a sufficient length of time to become saturated with dissolved oxygen. The water should be aerated by shaking a partially filled bottle or using a supply of clean compressed air. The distilled water used should be as near as possible to 20° C and of high purity. Place the desired volume of distilled water in a suitable bottle and add 1 mL each of phosphate buffer, magnesium sulfate, calcium chloride, and ferric chloride for each liter of water.

— Seeding material

Satisfactory seed may sometimes be obtained by using the supernatant liquor from domestic sewage that has been stored at 20° C for 24-36 h. Use the seed that has been found by practical experience to be the most satisfactory for the particular material under study. Only past experience can determine the amount of seed to be added per liter but the amount should give an oxygen depletion of approximately 2 mg/L. The amount of seed required may vary with the source of the seed. If the sample contains organic compounds not amenable to oxidation by domestic sewage seed, it may be necessary to use seed prepared from soil, an acclimated seed developed in the laboratory, or sediments collected below a particular waste discharge (preferably 2-5 mi below the point of discharge). Seeded dilution water should be used the same day it is prepared.

- Standards preparation

— Prepare a stock BOD standard solution by dissolving 0.150 g reagent grade glucose and 0.150 g reagent grade glutamic acid in 1 L of distilled water. The solids should be dried for 1 h at 103° C prior to weighing.

- Sample preparation

— Allow samples to warm to room temperature.

— Homogenize each sample mechanically.

— Remove a representative aliquot (approximately 25 g) and analyze for total solids content.

— Remove a representative aliquot (approximately 5 g) and weigh it to the nearest 0.1 mg.
— Transfer the weighed aliquot to a BOD bottle for analysis.

- Analytical procedures
  — Fill each BOD bottle with dilution water and place the samples in the incubator. Ensure that air bubbles are not trapped in the BOD bottles. Prepare a blank consisting of dilution water in a separate BOD bottle. Make sure that there is a water seal in the neck of each sample bottle and blank when placed in the incubator. Replenish the water seals on all bottles each morning.
  — Determine the initial dissolved oxygen concentration of each sample and blank using the Winkler titration method or a dissolved oxygen probe. This can best be accomplished by directly measuring the dissolved oxygen concentration in the dilution water. This method is recommended because sediment may cause a rapid consumption of oxygen, making it difficult to obtain a stable initial dissolved oxygen reading. If a probe is used for oxygen measurement, the same sample can be used for immediate dissolved oxygen demand and biochemical oxygen demand.
  — Incubate samples and blanks for 5 days at 20 ± 1° C. Determine residual dissolved oxygen concentrations in the incubated samples using the analytical method of choice. The most reliable BOD determinations will occur in those samples with a residual dissolved oxygen concentration of at least 2 mg/L and a dissolved oxygen depletion of at least 2 mg/L.
  — It is recommended that the dilution water be incubated as a check on its quality. To do this, fill two BOD bottles with unseeded dilution water. Stopper one bottle, fill the water seal, and place in the incubator at 20 ± 1° C for 5 days. Analyze the second sample to determine initial dissolved oxygen concentration. Following the 5-day period, determine dissolved oxygen in the incubated sample. The oxygen depletion should not be more than 0.2 mg/L and preferably not more than 0.1 mg/L. If these values are exceeded, the quality of the dilution water or the treatment of samples (e.g., filling of water seals) should be considered suspect.

  — Prepare a working BOD standard solution by diluting 20 mL of the stock solution to 1 L with seeded dilution water. Fill three BOD bottles and incubate at 20 ± 1° C for 5 days. The resulting BOD of these samples should be 218 ± 11 mg/L. Any appreciable deviation from these expected results may raise questions on the quality of the dilution water, the viability or suitability of the seed material, or the analytical technique.

- Calculations
  — Sediment BOD is calculated as follows:

$$\text{BOD (mg/kg dry weight)} = \frac{(O-F)\,(b)}{(g)\,(\%S)}$$

Where:

0 =	dissolved oxygen concentration at time zero, mg/L
F =	dissolved oxygen concentration after 5 days, mg/L
b =	volume of BOD bottle, mL
g =	wet weight of sediment sample used, g
%S =	percent solids in sediment sample (expressed as a decimal fraction.)

QA/QC Procedures

It is critical that each sample be thoroughly homogenized in the laboratory before a subsample is taken for analysis. Laboratory homogenization should be conducted even if samples were homogenized in the field.

It is recommended that triplicate analyses be conducted on one of every 20 samples, or on one sample per batch if less than 20 samples are analyzed. A dilution water blank and glucose-glutamic acid standard should be analyzed at the same frequency as the triplicate analyses.

DATA REPORTING REQUIREMENTS

Biochemical oxygen demand should be reported as mg/kg dry weight, to the nearest 0.1 unit. The laboratory should report the results of all samples analyzed, including QA replicates, seeded dilution water blanks, unseeded dilution water blanks, and glucose-glutamic acid standards. The laboratory should also note any problems that may have influenced sample quality.

CHEMICAL OXYGEN DEMAND (COD)

USE AND LIMITATIONS

Chemical oxygen demand 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 biochemical oxygen demand test for estimating organic matter. For samples from a specific source, chemical oxygen demand can be related empirically to biochemical oxygen demand, total organic carbon, or total volatile solids and then used for monitoring after a relationship has been established.

Major limitations of the chemical oxygen demand 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 chemical oxygen demand not be equated with organic matter in sediments.

FIELD PROCEDURES

Collection

Samples can be collected in glass or plastic containers. A minimum sample size of 50 g is recommended. If unrepresentative material is to be removed from the sample, it should be removed in the field under the supervision of the chief scientist and noted on the field log sheet.

Processing

Samples should be stored at 4° C and can be held under that condition for 7 days. Samples must be kept field-moist and free from air contact during storage to prevent air oxidation.

LABORATORY PROCEDURES

Analytical Procedures

- Equipment
  - Reflux apparatus
    Consisting of 250- or 500-mL Erlenmeyer flasks with ground glass 24/40 neck[1] and 300-mm jacket Liebig, West, or equivalent condensers[2] with 24/40 ground glass joint.
  - Hot plate

[1]Corning 5000 or equivalent.
[2]Corning 2360, 91548, or equivalent.

Having sufficient power to produce 1.4 W/cm^2 (9 W/in^2) of heating surface, or equivalent, to ensure adequate refluxing of the sample.

— Standard potassium dichromate solution, 0.250 N
  Dissolve 12.259 g $K_2Cr_2O_7$ primary standard grade, previously dried at 103° C for 2 h, in distilled water and dilute to 1 L. The addition of 0.12 g/L sulphamic acid will eliminate interference due to nitrites in the sample at concentrations up to 6 mg/L.
— Sulfuric acid reagent
  Concentrated H_2SO_4 containing 22 g silver sulfate, Ag_2SO_4, per 9-lb bottle. Allow 1 or 2 days for dissolution.
— Standard ferrous ammonium sulfate titrant, 0.25 N
  Dissolve 98 g $Fe(NH_4)_2 (SO_4)_2 \cdot 6H_2O$ in distilled water. Carefully add 20 mL concentrated H_2SO_4, cool, and dilute to 1 L. This solution must be standardized against $K_2Cr_2O_7$ daily. To standardize the ferrous ammonium sulfate, dilute 10 mL standard potassium dichromate solution to approximately 100 mL. Carefully add 30 mL concentrated H_2SO_4 and allow to cool. Titrate with ferrous ammonium titrant, using 2-3 drops of ferroin indicator.

  $$\text{Normality} = \frac{(\text{mL } K_2Cr_20_7)\ (0.25)}{[\text{mL Fe } (NH_4)2\ (S0_4)_2]}$$

— Ferroin indicator
  Dissolve 1.485 g 1,10-phenantroline monohydrate and 0.695 g ferrous sulfate, $FeSO_4 \cdot 7H_2O$, in water and dilute to 100 mL. Alternatively, a commercially prepared indicator can be purchased.
— Silver sulfate
  Ag_2SO_4, reagent powder.
— Mercuric sulfate
  $HgSO_4$, analytical-grade crystals.

- Sample preparation
  — Allow samples to warm to room temperature.
  — Homogenize each sample mechanically.
  — Remove a representative aliquot (approximately 25 g) and analyze for total solids content.
  — Remove a representative aliquot (approximately 2 g) and weigh it to the nearest 0.1 mg.
  — Transfer the weighed aliquot to a reflux flask for COD analysis. Wash the sediment into the flask with a minimum amount of distilled water (i.e., <25 mL).

- Analytical procedures
  — Place several boiling stones or glass beads and 1.0 g $HgSO_4$ in the reflux flask with the sample.

— Add 25 mL 0.25 N $K_2CR_2O_7$ to the flask and mix thoroughly.
— Slowly, and with constant mixing, add 75 mL of sulfuric acid-silver sulfate solution. Ensure that the mixture is well mixed to avoid localized superheating.
— Attach the sample flask to a condenser and reflux for 2 h. If the added dichromate dissipates during reflux, either 1) repeat, using a smaller sample size, or 2) carefully add additional 0.25 N $K_2Cr_2O_7$ through the condenser. Be sure to record any added dichromate.
— Allow the sample to cool and rinse the condenser with 40-50 mL distilled water.
— Add an additional 50 mL of distilled water to the sample and allow to cool to room temperature.
— Add 3-5 drops of ferroin indicator and titrate with 0.25 N $Fe(NH_4)_2(SO_4)_2$ to a sharp color change (blue-green to reddish-brown).
— For a blank, reflux 25 mL of distilled water, 25 mL of 0.25 N $K_2Cr_2O_7$, 1 g $HgSO_4$, several glass beads or boiling stones, and 75 mL of sulfuric acid-silver sulfate solution for 2 h. Cool, add 3-5 drops of ferrion indicator, treat as a sample and titrate with 0.25 N $Fe(NH_4)_2(SO_4)_2$.

• Calculations
— Sediment COD is determined as follows:

$$\text{COD (mg/kg dry weight)} = \frac{(A-B)\,(N)\,(8{,}000)}{(g)\,(\%S)}$$

Where:

A = volume of 0.25 N $Fe(NH_4)_2(SO_4)_2$ for blank titration, mL
B = volume of 0.25 N $Fe(NH_4)_2(SO_4)_2$ for sample titration, mL
N = normality of 0.25 N $Fe(NH_4)_2(SO_4)_2$ used for titration, eq/L
8,000 = equivalent weight of oxygen, mg/eq
g = wet weight of sample, g
%S = percent solids in sediment sample (expressed as a decimal fraction.

QA/QC Procedures

It is critical that each sample be thoroughly homogenized in the laboratory before a subsample is taken for analysis. Laboratory homogenization should be conducted even if samples were homogenized in the field.

It is recommended that triplicate analyses be conducted on one of every 20 samples, or on one sample per batch if less than 20 samples are analyzed.

DATA REPORTING REQUIREMENTS

Chemical oxygen demand should be reported as mg/kg of sediment dry weight to the nearest 0.1 unit. The laboratory should report the results of all samples analyzed (including QA replicates and method blanks) and should note any problems that may have influenced data quality. The laboratory should also report the results of the standard test.

REFERENCES

Buchanan, J.B. 1984. Sediment analysis. pp. 41-65. In: Methods for the Study of Marine Benthos. N.A. Holme and A.D. McIntyre (eds). Blackwell Scientific Publications, Boston, MA.

Folk, R.L. 1968. Petrology of sedimentary rocks. Hemphill Publishing Co., Austin, TX. 172 pp.

Hedges, J.I., and J.H. Stern. 1984. Carbon and nitrogen determinations of carbonate-containing solids. Limnol. Oceanogr. 29:657-663.

Krunbein, W.C. and F.J. Pettijohn. 1938. Manual of sedimentary petrography. Appleton-Century Crafts, NY. 549 pp.

Plumb, R.H. 1981. Procedures for handling and chemical analysis of sediment and water samples. Technical Report EPA/CE-81-1. U.S. Army Corps of Engineers, Vicksburg, MS.

U.S. Environmental Protection Agency. 1983. Methods for chemical analysis of water and waste. EPA 600/4-79-020 (Revised 1983). Environmental Monitoring and Support Laboratory, Cincinnati, OH.