

Field Sampling Manual  
for the  
Regional Monitoring Program for Trace Substances

Prepared for the  
San Francisco Estuary Institute

by

Applied Marine Sciences  
4749 Bennett Drive  
Suite L  
Livermore, CA 94550

## ACKNOWLEDGEMENTS

This document was produced with the assistance from the following  
Regional Monitoring Program participants:

Russell Flegal and Genine Scelfo  
Institute of Marine Sciences  
University of California at Santa Cruz

John Hunt and Bryn Phillips  
Marine Pollution Studies

Wally Jarman Jocelyn Vedder  
Energy and Geoscience Institute  
University of Utah

Mike Kellogg  
City and County of San Francisco

## TABLE OF CONTENTS

1	PROGRAM OVERVIEW .....	1
1.1	RMP Rationale.....	1
1.2	RMP Objectives .....	1
1.3	RMP Organization and Management.....	2
1.3.1	Telephone and E-mail Contact List .....	4
2	CRUISE SCHEDULING.....	5
2.1	Water.....	5
2.2	Sediment .....	5
2.3	Bioaccumulation .....	7
2.4	Site Data.....	7
3	SAMPLING METHODS.....	10
3.1	Water Sampling .....	10
3.1.1	Overview and Objectives.....	10
3.1.2	Water Sampling Vessel Safety.....	11
3.1.3	Water Sampling Equipment List.....	13
3.1.4	Sample Containers .....	16
3.1.5	Trace Elements Sampling Equipment Preparation .....	16
3.1.6	Trace Organics Sampling Equipment Preparation.....	22
3.1.7	CTD Preparation .....	22
3.1.8	Water Sampling Procedures.....	24
3.1.8.1	Water Trace Metals Sampling .....	24
3.1.8.2	General Water Quality Measurements.....	25
3.1.9	Water Organics Sampling .....	25
3.1.10	Water CTD Profiling.....	28
3.1.11	Water Toxicity Sampling .....	29
3.1.12	Watershed Water Sampling .....	30
3.1.13	Water Sample Handling and Shipping.....	31
3.2	Sediment Sampling .....	33
3.2.1	Overview and Objectives.....	33
3.2.2	Sediment Sampling Vessel Safety .....	34
3.2.3	Sediment Sampling Equipment List .....	35
3.2.4	Sample Containers .....	36
3.2.5	Sediment Sampling Equipment Preparation .....	37
3.2.6	Sediment Sampling Procedures .....	38
3.2.6.1	Sediment Chemistry.....	39
3.2.6.2	Benthic Infauna.....	39
3.2.6.3	Sediment Pore Water Analysis .....	41
3.2.6.4	Sediment CTD Profiling .....	45
3.2.6.5	Watershed Sediment Sampling .....	45
3.2.6.6	Sediment Sample Storage and Handling.....	45
3.3	Bioaccumulation Sampling.....	47
3.3.1	Overview and Objectives.....	47

3.3.2	Oyster Collection .....	47
3.3.3	Mussel Collection .....	48
3.3.4	Resident Clam Collection .....	48
3.3.5	Vessel Safety.....	49
3.3.6	Dive Safety.....	49
3.3.6.1	Dive Team Member Responsibilities.....	50
3.3.6.2	Dive Equipment Use and Maintenance.....	50
3.3.7	Dive Operations .....	51
3.3.7.1	Dive Records.....	52
3.3.7.2	Equipment.....	52
3.3.8	Dive Equipment Preparation.....	53
3.3.9	Bioaccumulation Sample Handling .....	53
3.3.10	Bivalve Handling and Storage .....	54
3.3.11	Bivalve Chain of Custody.....	55
4	APPENDIX A. FIGURES .....	56

## TABLES

Table 1. Principal Investigators of the RMP.....	3
Table 2. Telephone and e-mail contacts for RMP researchers and staff. ....	4
Table 3. Water cruise activity schedule. ....	5
Table 4. Sediment cruise activity schedule.....	6
Table 5. Bioaccumulation cruise activity schedule.....	7
Table 6. Site name, codes and coordinates of RMP sample locations.....	8
Table 7. Crew responsibilities for RMP water sampling cruise. ....	11
Table 8. Equipment list for water trace elements sampling.....	13
Table 9. Equipment list for water trace organics sampling. ....	15
Table 10. Miscellaneous support equipment list for water toxicity sampling. ....	16
Table 11. Container list for water sampling.....	16
Table 12. Crew responsibilities for RMP sediment cruise. ....	33
Table 13. Equipment list for sediment sampling. ....	36
Table 14. Container list for sediment sampling.....	37
Table 15. Equipment list for pore water sampling.....	42
Table 16. Storage methods for sediment samples.....	46
Table 17. Dive operations task list.....	52
Table 18. Equipment list for bivalve sampling cruise. ....	53

# **1 Program Overview**

## **1.1 RMP Rationale**

The Sacramento-San Joaquin Delta and the San Francisco Bay (together known as the Estuary) make up the West Coast's largest estuary. The watershed that drains into the 1,600 mi<sup>2</sup> Estuary comprises over 40% of California's surface area. The Estuary is in close proximity to and highly affected by both a highly urbanized landscape and the rich agricultural areas of the Central Valley. Urban runoff, agricultural runoff, treated wastewater, and dredging activities all introduce contaminants to estuarine waters.

Scientists have been conducting research and monitoring activities on the Estuary for decades. These activities were typically geared for a specific need, limited in coverage, and failed to provide an overall picture of the Estuary's condition. In addition, sampling methods used by different studies were rarely comparable.

The San Francisco Bay Regional Water Quality Control Board (Regional Board) is the state agency responsible for implementing and overseeing water quality programs for the San Francisco Estuary. The Regional Board realized the need for comprehensive long-term monitoring and established the Regional Monitoring Program for Trace Substances (RMP) in an effort to fulfill that need. In addition to the Regional Board, two other entities help determine the focus and operations of the RMP: the Steering Committee, made up of representatives of the seventy-seven public and private organizations that discharge treated wastewater, cooling water, or urban runoff, and the San Francisco Estuary Institute (SFEI) who is responsible for implementation of the RMP.

The RMP regularly monitors contaminant concentrations in water, sediments, and fish and shellfish tissue in the Estuary. This contaminant monitoring allows the Regional Board to evaluate the effectiveness of its water quality programs in its overarching goal of protecting the beneficial uses of the Estuary.

## **1.2 RMP Objectives**

The RMP's overall goal is to provide data and interpretation that helps to address certain of the Regional Board's information needs. These efforts fall under five major objectives:

- Describe patterns and trends in contaminant concentration and distribution.
- Describe general sources and loadings of contamination to the Estuary.
- Measure contaminant effect on selected parts of the Estuary ecosystem.
- Compare monitoring information to relevant water quality objectives and other guidelines.
- Synthesize and distribute information from a range of sources to present a more complete picture of the sources, distribution, fates, and effects of contaminants in the Estuary ecosystem.

### 1.3 RMP Organization and Management

The organization diagrams for water, sediment and bioaccumulation sampling are shown in Appendix A, Figures 1, 2 and 3, respectively. Responsibilities for each cruise are as follows:

For water sampling, Applied Marine Sciences (AMS) is responsible for drafting the sample cruise plan in accordance with the goals of SFEI and the Steering Committee, conducting CTD casts and collecting samples for water toxicity. The University of California, Santa Cruz (UCSC) is responsible for trace elements (also referred to as trace metals) water sample collection and analysis/reporting of trace elements and cognates data. UCSC also collects samples for analysis of As, Hg, and Se that are shipped by AMS for analysis and reporting by Brooks Rand, Ltd. (BRL). The University of Utah (UU) is responsible for collecting, analyzing and reporting all trace organics water samples. The *R/V David Johnston*, a UCSC-operated vessel, is the vessel used for conducting this sampling.

For sediment sampling, AMS is responsible for generating the cruise plan in accordance with the goals of SFEI and the Steering Committee. AMS is also responsible for sample collection, sample processing, pore water analysis of pH and ammonia, CTD profiling, and sample distribution. SFEI is responsible for assisting with sample collection and processing in the field and for overall data compilation and analysis. The *R/V David Johnston*, a UCSC vessel, is also operated by UCSC personnel. The Bay Area Dischargers Authority (BADA) is responsible for analysis and reporting of trace elements and trace organics samples. Texas A&M Geochemical and Environmental Research Group (GERG) is responsible for analysis and reporting of trace organics intercalibration samples. The Marine Pollution Studies Laboratory (MPSL) is responsible for analysis and reporting of sediment toxicity. UCSC is responsible for analysis and reporting of cognates and intercalibration analysis and reporting of trace elements samples. BRL is responsible for analysis and reporting of As, Hg, and Hg samples. The City and County of San Francisco are responsible for collection of benthic infauna samples and analysis and reporting of the results. Finally, the United States Geological Survey (USGS) is responsible for analysis of foraminifera samples. USGS data are not part of the official RMP data program.

For bioaccumulation sampling, AMS is responsible for generating the cruise plan in accordance with the goals of SFEI and the Steering Committee. AMS is also responsible for acquisition of all bivalves prior to the cruises; sample deployment, maintenance and retrieval via SCUBA diving; homogenization of bivalve samples for analysis of trace elements; condition index analysis; and additionally, vessel supply and operations for the Davis Point site (BD40). The vessel used for all RMP sites, except Davis Point, is the *R/V Questuary*, supplied and operated by the Romberg Tiburon Center. BADA is responsible for analysis and reporting of tissue trace elements and trace organics. GERG is responsible for homogenizing trace organics samples and intercalibration samples for trace organics. BRL is responsible for analysis and reporting of As, Hg, and Hg. UCSC is responsible for intercalibration of trace elements. SFEI is responsible for overall data compilation and analysis.

The principal investigators of the RMP are listed in Table 1.

**Table 1. Principal Investigators of the RMP.**

<i>Contractor</i>	<i>Affiliation</i>
Prime Contractors	Dr. Bob Spies and Dr. Andy Gunther Applied Marine Sciences (AMS), Inc., Livermore, CA
Trace Element Chemistry	Dr. Russ Flegal, UC Santa Cruz (UCSC), CA ; Dr. Eric Prestbo, Brooks-Rand, Ltd., (BRL) Seattle, WA
Trace Organic Chemistry	Dr. Terry Wade, Texas A&M University (GERG), TX; Dr. Walter Jarman, University of Utah, CA
Water Toxicity Testing	Dr. Scott Ogle Pacific Eco-Risk Laboratories (PERL), Martinez CA
Sediment Toxicity Testing	Mr. John Hunt and Mr. Brian Anderson Marine Pollution Studies Lab (MPSL), Granite Canyon, CA
Bagged Bivalve Sampling	Dr. Andy Gunther, AMS, Livermore, CA
USGS Water Quality	Dr. James Cloern, US Geological Survey (USGS), Menlo Park, CA
USGS Sediment Transport	Dr. David Schoellhamer, USGS, Sacramento, CA
Pilot Study on Benthic Macrofauna	Dr. Bruce Thompson San Francisco Estuary Institute (SFEI), Richmond, CA; Ms. Heather Peterson Dept. of Water Resources, Sacramento, CA
Watershed Pilot Study	Dr. Rainer Hoenicke, SFEI, Richmond, CA

### 1.3.1 Telephone and E-mail Contact List

The telephone numbers and e-mail contacts of RMP investigators and staff are presented in Table 2.

**Table 2. Telephone and e-mail contacts for RMP researchers and staff.**

<i>Last</i>	<i>First</i>	<i>Affiliation</i>	<i>Phone</i>	<i>E-mail</i>
Anderson	Brian	Marine Pollution Studies Lab	831 624 0947	<a href="mailto:bsanders@cats.ucsc.edu">bsanders@cats.ucsc.edu</a>
Andrade-Bunnell	Jo	City of San Jose	408 945 3716	<a href="mailto:jo.andrade-bunnell@ci.sj.ca.us">jo.andrade-bunnell@ci.sj.ca.us</a>
Bacon	Corinne	University of Utah, Energy/Geoscience Inst.	801 585 9178	<a href="mailto:cbacon@egi.utah.edu">cbacon@egi.utah.edu</a>
Bell	David	Applied Marine Sciences	925 373 7142	<a href="mailto:bell@amarine.com">bell@amarine.com</a>
Daum	Ted	San Francisco Estuary Institute	510 231 9539	<a href="mailto:ted@sfei.org">ted@sfei.org</a>
Flegal	Russ	University of California	831 459 2093	<a href="mailto:rflegal@emerald.ucsc.edu">rflegal@emerald.ucsc.edu</a>
Gold	Jordan	Applied Marine Sciences	925 373 7142	<a href="mailto:gold@amarine.com">gold@amarine.com</a>
Gunther	Andrew	Applied Marine Sciences	925 373 7142	<a href="mailto:gunther@amarine.com">gunther@amarine.com</a>
Hardin	Dane	Applied Marine Sciences	831 426 6326	<a href="mailto:hardin@amarine.com">hardin@amarine.com</a>
Hoenicke	Rainer	San Francisco Estuary Institute	510 231 9539	<a href="mailto:rainer@sfei.org">rainer@sfei.org</a>
Hunt	John	University of California, Santa Cruz	831 624 0947	<a href="mailto:jhunt@hydrogen.ucsc.edu">jhunt@hydrogen.ucsc.edu</a>
Jarman	Wally	University of Utah, Energy/Geoscience Inst.	801 585 3082	<a href="mailto:wjarman@egi.utah.edu">wjarman@egi.utah.edu</a>
Johnson	Jay	Applied Marine Sciences	925 373 7142	<a href="mailto:johnson@amarine.com">johnson@amarine.com</a>
Kellogg	Michael	City & County of San Francisco	415 242 2218	<a href="mailto:mkellogg@ix.netcom.com">mkellogg@ix.netcom.com</a>
Kist	Bill	Brooks-Rand, Ltd.	206 632 6206	
Lowe	Sarah	San Francisco Estuary Institute	510 231 9539	<a href="mailto:sarah@sfei.org">sarah@sfei.org</a>
McGann	Mary	United States Geological Service	650 329 4979	
Morgan	David	Romberg Tiburon Center	415 435 7123	<a href="mailto:rvquest@aol.com">rvquest@aol.com</a>
Ogle	Scott	Pacific Eco-Risk Laboratories	510 313 8080	<a href="mailto:scottogle@eco-risk.com">scottogle@eco-risk.com</a>
Phillips	Bryn	Marine Pollution Studies Lab	831 624 0947	<a href="mailto:bnp@cats.ucsc.edu">bnp@cats.ucsc.edu</a>
Salop	Paul	Applied Marine Sciences	925 373 7142	<a href="mailto:salop@amarine.com">salop@amarine.com</a>
Scelfo	Genine	University of California	831 459 3563	<a href="mailto:gscelfo@emerald.ucsc.edu">gscelfo@emerald.ucsc.edu</a>
Smith	Gordon	University of California	831 459 4735	<a href="mailto:rvboat@ucsc.edu">rvboat@ucsc.edu</a>
Spies	Robert	Applied Marine Sciences	925 373 7142	<a href="mailto:spies@amarine.com">spies@amarine.com</a>
Targart	Laura	City & County of San Francisco	415 242 2218	
Tenbrook	Patti	East Bay Municipal Utility District		<a href="mailto:ptenbrook@ebmud.com">ptenbrook@ebmud.com</a>
Vedder	Jocelyn	University of Utah, Energy/Geoscience Inst.	801 585 9178	<a href="mailto:jvedder@egi.utah.edu">jvedder@egi.utah.edu</a>
Vwpiekarski	Vitek	Marine Pollution Studies Lab	831 624 0947	<a href="mailto:vwpiekars@cats.ucsc.edu">vwpiekars@cats.ucsc.edu</a>

## 2 Cruise Scheduling

### 2.1 Water

The typical water cruise activity schedule assumes that an average of 1.5 hours is required for sampling each station and that the survey vessel is capable of maintaining a minimum cruising speed between stations of 7 knots. Actual survey times will vary depending on weather and sampling conditions. A typical cruise schedule is presented in Table 3.

**Table 3. Water cruise activity schedule.**

<i>Day</i>	<i>Activity Schedule</i>
Day 1	Mobilize gear on vessel R/V David Johnston at the Emeryville Marina. Conduct safety briefing and depart for South Bay sites. Sample Oyster Point, San Bruno Shoal, Coyote Creek, Redwood Creek, and South Bay. Transit to Redwood City.
Day 2	Mobilize gear on vessel R/V David Johnston. Depart Redwood City, sample Dumbarton Bridge, San Jose, and Sunnyvale sites. Transit to Emeryville Marina.
Day 3	Mobilize gear on vessel R/V David Johnston. Depart Emeryville Marina, sample Yerba Buena Island, Alameda, Golden Gate and Richardson Bay sites. Transit to Emeryville Marina.
Day 4	Mobilize gear on vessel R/V David Johnston. Depart Emeryville Marina, sample Red Rock and Point Isabel sites. Transit to Emeryville Marina. As, Se, Hg samples are shipped to Brooks Rand.
Day 5	R/V David Johnston transits to Martinez Marina. No crew is required.
Day 6	Mobilize gear on vessel R/V David Johnston. Depart Martinez Marina, sample Davis Point, Pinole Point, San Pablo Bay, and Petaluma River sites. Transit to Martinez Marina.
Day 7	Mobilize gear on vessel R/V David Johnston. Depart Martinez Marina, sample Honker Bay, Napa river, and Pacheco Creek sites. Transit to Martinez Marina.
Day 8	Mobilize gear on vessel R/V David Johnston. Depart Martinez Marina, sample Grizzly Bay, Sacramento, and San Joaquin river sites. Transit to Martinez Marina and demobilize gear off vessel.
Day 9	Sample watershed sites at Standish Dam (Coyote Creek) and South Bay Yacht Club (Guadalupe River). As, Se, Hg samples are shipped to Brooks Rand.

### 2.2 Sediment

The typical sediment cruise activity schedule assumes that benthic sampling requires approximately 0.25 hours/grab/site, and that the survey vessel is capable of maintaining a cruising speed of 7 knots between stations during which time benthic samples are processed. A typical sediment cruise schedule is presented in Table 4.

**Table 4. Sediment cruise activity schedule.**

<i>Day</i>	<i>Activity Schedule</i>
Day 1	<p>Mobilize gear on vessel R/V David Johnston, conduct safety briefing at the Martinez Marina. Depart for Grizzly Bay.</p> <p>Sample at Grizzly Bay, Honker Bay, Sacramento River, San Joaquin River, and Pacheco Creek. Drop off crew at Martinez Marina and shuttle cars to Vallejo. Vessel transits to Vallejo Marina.</p> <p>Demobilize gear at Vallejo Marina. Load benthic sampling gear onto vessel. All chemistry and toxicity samples will be stored on ice aboard the vessel.</p>
Day 2	<p>Mobilize gear on vessel R/V David Johnston at the Vallejo Marina. Benthic sampling crew member joins the rest of the sampling crew. Depart for Napa River.</p> <p>Sample at Napa River, Davis Point, Petaluma River, San Pablo Bay, Pinole Point, and Red Rock. Vessel transits to the Emeryville Marina.</p> <p>Demobilize gear at the Emeryville Marina. Toxicity samples are stored on wet ice aboard the vessel. Pore water samples are stored in a light-proof container. Trace elements, trace organics, As, Hg, Se and cognate samples are stored on dry ice aboard the vessel. Foraminifera samples are stored on deck in a leak-proof container. Benthic samples are removed from the vessel by CCSF.</p> <p>The crew shuttles back to Vallejo to retrieve their cars.</p>
Day 3	<p>Mobilize gear on vessel R/V David Johnston at the Emeryville Marina. Depart for Point Isabel.</p> <p>Sample at Point Isabel, Richardson Bay, Horseshoe Bay, and Yerba Buena Island. Vessel transits back to Emeryville.</p> <p>Demobilize gear at Emeryville Marina. All chemistry, benthic, and toxicity samples will be stored on ice aboard the vessel.</p>
Day 4	<p>Mobilize gear on vessel R/V David Johnson at the Emeryville Marina. Depart for Alameda.</p> <p>Sample at Alameda, Oyster Point, San Bruno Shoal, Redwood Creek, and South Bay. Vessel transits to Redwood City USGS dock.</p> <p>Demobilize gear at Redwood City. Unload benthic sampling gear. Benthic samples will be removed from the vessel by CCSF. All chemistry and toxicity samples will be stored on ice aboard the vessel.</p>
Day 5	<p>Mobilize gear on vessel R/V David Johnston at the Redwood City USGS dock. Depart for Dumbarton Bridge.</p> <p>Sample at Dumbarton Bridge, San Jose, Sunnyvale and Coyote Creek. Vessel transits to Emeryville.</p> <p>Demobilize gear at Emeryville Marina. All toxicity and pore water samples are picked up at the vessel by a representative from MPSL. Trace metal and organic samples are picked up at the vessel by a BADA representative. As, Hg, Se samples for BRL are transferred to AMS for shipment the following day. Foram samples are stored at AMS until pick-up by USGS personnel.</p>
Day 6	<p>Sample Standish Dam and Guadalupe River. Send all remaining samples to the proper analytical laboratories.</p>

### 2.3 Bioaccumulation

Bioaccumulation cruises are conducted for deployment of bivalves, maintenance of bivalves approximately 50 days after deployment and retrieval of bivalves approximately 100 days after deployment. The typical bioaccumulation deployment cruise activity schedule assumes that dive operations require approximately 0.5 hours on site and that the diver support vessel is capable of maintaining a cruising speed of 12 knots between sites. A typical bioaccumulation cruise schedule is presented in Table 5.

**Table 5. Bioaccumulation cruise activity schedule.**

<i>Day</i>	<i>Activity</i>
Day 1	Approximately 7-14 days prior to the start of a deployment cruise, oysters ( <i>C. gigas</i> ) are obtained from a commercial grower located in Tomales Bay. Oysters are transferred from the grower's tanks into mesh bags and placed into a holding tank at the Bodega Marine Lab (BML). Normally during the same day, mussels ( <i>M. californianus</i> ) are collected during low tides from Bodega Head, Sonoma County. Mussels are transferred into mesh bags and also placed into a holding tank at BML. The holding tank at the BML is constantly flushed with filtered seawater at ambient ocean temperatures. Bivalves are kept in holding tanks no less than 24 hours and no longer than 14 days prior to deployment.
Day 2	All bivalves are retrieved from the holding tank at the BML. (This task occurs within 2-14 days after bivalves are collected)
Day 3	Mobilize gear on vessel M.E. II, conduct safety briefing at Martinez Marina. Deploy bivalves at Davis Point. Demobilize gear at Martinez Marina.
Day 4	Mobilize gear on vessel R/V Questuary, conduct safety briefing at Emeryville Marina. Deploy bivalves at Yerba Buena Island, Alameda, Redwood Creek, Dumbarton Bridge and Coyote Creek. Demobilize gear at Emeryville Marina, refill SCUBA tanks as necessary.
Day 5	Mobilize gear on vessel R/V Questuary, Emeryville Marina. Deploy bivalves at Horseshoe Bay, Red Rock, Pinole Point, San Pablo Bay, Petaluma River, and Napa River. Demobilize gear at Martinez Marina.
Day 6	Mobilize gear on vessel R/V Questuary, Martinez Marina. Deploy Bivalves at Grizzly Bay. Conduct clam harvesting operations at Sacramento river and San Joaquin river as needed and return to Emeryville Marina. Demobilize gear.

### 2.4 Site Data

As one objective of the RMP is to determine seasonal and annual trends in chemical and biological water quality, it is important to maintain sample sites that are as far as possible from the influence of major contaminant sources. In this way, temporal and spatial variability in analytical data can be interpreted without the confounding influence of variable contaminant input from nearby sources. This criterion requires that stations be located in places of higher dilution, such as close to the major channels in each embayment.

Another objective of the RMP is to determine the spatial distribution of contamination in the Estuary. This requires that stations be located throughout the Estuary, from the extreme

South Bay all the way up the northern reach of the Estuary to the mouths of the Sacramento and San Joaquin rivers. This also requires that samples be collected approximately the same time each year, so that data may be compared over several years during the same seasonal period.

Site selection is also influenced by those stations monitored in the Estuary during the pilot phase of the RMP, conducted by the Regional Board from 1989 through 1992. Locating RMP sample sites at or near pilot sites provides a larger database that is directly comparable to the data collected during the pilot program. This is because many of the analytical and sampling methods that were used in the pilot phase of the program have been adopted for use in the current RMP.

Finally, many sample sites are chosen due to methodological requirements in the field. For example, normal water sampling operations requires the vessel to remain at a site for at least an hour without the engines running. This means that stations cannot be located in a vessel traffic lane, which is a frequent use of estuarine channels. Such feasibility requirements have been addressed in the pilot program, and this knowledge was applied in selecting the sampling sites for the RMP.

It is the responsibility of the vessel captain to ensure that the vessel reaches and maintains the proper location for each sample site. It is the responsibility of the cruise manager to verify the accuracy of sample site coordinates and to record the coordinates in a cruise logbook. This logbook contains the time of arrival, time of departure, latitude and longitude (measured from vessel's global positioning system) and general sea conditions if outside the normal range of conditions for the station being sampled. Sampling coordinates are checked throughout sampling to ensure that the anchor has not dragged. For sediment sampling, coordinates are recorded for each replicate grab at each sampling location. The site names, codes and coordinates for all RMP sample locations (water, sediment and bioaccumulation deployments and collections) are listed in Table 6.

**Table 6. Site name, codes and coordinates of RMP sample locations.**

<i>Site Name</i>	<i>Code</i>	<i>Sample Matrix</i>	<i>Measurements Made</i>	<i>Sample Events</i>	<i>Latitude</i>		<i>Longitude</i>	
					deg	min	deg	min
Coyote Creek	BA10	water	Q,M,O,T	3	37	28.20'	122	03.80'
	BA10	sediment	Q,M,O	2	37	28.20'	122	03.80'
	BA10	bioaccumulation	M,O,C	2	37	28.19'	122	03.83'
South Bay	BA20	water	Q,M	3	37	29.69'	122	05.34'
	BA21	sediment	Q,M,O,T	2	37	29.64'	122	05.25'
Dumbarton Bridge	BA30	water	Q,M,O	3	37	30.90'	122	08.11'
	BA30	sediment	Q,M,O	2	37	30.87'	122	08.08'
	BA30	bioaccumulation	M,O,C	2	37	30.80'	122	08.08'
Redwood Creek	BA40	water	Q,M,O,T	3	37	33.67'	122	12.57'
	BA41	sediment	Q,M,O,T	2	37	33.67'	122	12.62'
	BA40	bioaccumulation	M,O,C	2	37	32.82'	122	11.70'

Site Name	Code	Sample Matrix	Measurements Made	Sample Events	Latitude		Longitude	
					deg	min	deg	min
San Bruno Shoal	BB15	water	Q,M	3	37	37.00'	122	17.00'
	BB15	sediment	Q,M,O,T	2	37	37.00'	122	17.00'
Oyster Point	BB30	water	Q,M	3	37	40.20'	122	19.75'
	BB30	sediment	Q,M,O	2	37	40.21'	122	19.77'
Alameda	BB70	water	Q,M,O,T	3	37	44.66'	122	19.30'
	BB70	sediment	Q,M,O,T	2	37	44.84'	122	19.40'
	BB71	bioaccumulation	M,O,C	2	37	41.73'	122	20.38'
Yerba Buena Island	BC10	water	Q,M,O,T	3	37	49.36'	122	20.96'
	BC11	sediment	Q,M,O,T	2	37	49.44'	122	20.93'
	BC10	bioaccumulation	M,O,C	2	37	49.12'	122	20.81'
Golden Gate	BC20*	water	Q,M,O	3	.	.	.	.
Horseshoe Bay	BC21	sediment	Q,M,O,T	2	37	49.98'	122	28.43'
	BC21	bioaccumulation	M,O,C	2	37	49.87'	122	28.65'
Richardson Bay	BC30	water	Q,M	3	37	51.81'	122	28.66'
	BC32	sediment	Q,M,O	2	37	51.82'	122	28.72'
Point Isabel	BC41	water	Q,M	3	37	53.30'	122	20.55'
	BC41	sediment	Q,M,O	2	37	53.34'	122	20.55'
Red Rock	BC60	water	Q,M,O,T	3	37	55.00'	122	26.00'
	BC60	sediment	Q,M,O,T	2	37	55.00'	122	25.97'
	BC61	bioaccumulation	M,O,C	2	37	55.70'	122	28.13'
Petaluma River	BD15	water	Q,M,O,T	3	38	06.66'	122	29.00'
	BD15	sediment	Q,M,O	2	38	06.66'	122	29.00'
	BD15	bioaccumulation	M,O,C	2	38	06.77'	122	30.05'
San Pablo Bay	BD20	water	Q,M,O	3	38	02.92'	122	25.19'
	BD22	sediment	Q,M,O	2	38	02.86'	122	25.24'
	BD20	bioaccumulation	M,O,C	2	38	02.72'	122	25.71'
Pinole Point	BD30	water	Q,M,O,T	3	38	01.48'	122	21.65'
	BD31	sediment	Q,M,O,T	2	38	01.49'	122	21.71'
	BD30	bioaccumulation	M,O,C	2	38	01.00'	122	22.05'
Davis Point	BD40	water	Q,M,O	3	38	03.12'	122	16.62'
	BD41	sediment	Q,M,O,T	2	38	03.11'	122	16.65'
	BD40	bioaccumulation	M,O,C	2	38	03.26'	122	15'.63
Napa River	BD50	water	Q,M,O,T	3	38	05.79'	122	15.61'
	BD50	sediment	Q,M,O,T	2	38	05.79'	122	15.61'
	BD50	bioaccumulation	M,O,C	2	38	04.84'	122	14.82'
Pacheco Creek	BF10	water	Q,M	3	38	03.09'	122	05.80'
	BF10	sediment	Q,M,O	2	38	02.85'	122	05.66'
Grizzly Bay	BF20	water	Q,M,O,T	3	38	06.96'	122	02.31'
	BF21	sediment	Q,M,O,T	2	38	06.97'	122	02.35'
	BF20	bioaccumulation	M,O,C	2	38	06.49'	122	03.37'
Honker Bay	BF40	water	Q,M	3	38	04.00'	121	56.00'
	BF40	sediment	Q,M,O	2	38	04.00'	121	56.00'
Sacramento River	BG20	water	Q,M,O,T	3	38	03.56'	121	48.59'
	BG20	sediment	Q,M,O,T	2	38	03.36'	121	48.63'
	BG20	bioaccumulation	M,O,C	2	38	03'.58	121	47.50'

<i>Site Name</i>	<i>Code</i>	<i>Sample Matrix</i>	<i>Measurements Made</i>	<i>Sample Events</i>	<i>Latitude</i>		<i>Longitude</i>	
					deg	min	deg	min
San Joaquin River	BG30	water	Q,M,O,T	3	38	01.40'	121	48.45'
	BG30	sediment	Q,M,O,T	2	38	01.36'	121	48.44'
	BG30	bioaccumulation	M,O,C	2	38	01.27'	121	48.32'
San Jose	C-3-0	water	Q,M,T	3	37	27.85'	122	01.60'
	C-3-0	sediment	Q,M	2	37	27.72'	121	58.53'
Sunnyvale	C-1-3	water	Q,M,T	3	37	26.08'	122	00.64'
	C-1-3	sediment	Q,M	2	37	26.13'	122	00.67'

**Table 6 notes:**

<i>Abbreviation</i>	<i>Notes</i>
*	Location dependent on salinity
Q	Water and/or sediment quality
O	Trace organics
C	Bivalve condition index
M	Trace elements
T	Toxicity
P	Porewater chemistry

### 3 Sampling Methods

#### 3.1 Water Sampling

##### 3.1.1 Overview and Objectives

Water sampling for the RMP consists of sampling 26 stations in the Estuary, including two watershed sites located in the South Bay. A cruise normally requires seven to eight working days to complete, including one day for the vessel to transit between ports in Emeryville and Martinez. Water samples are collected in the total (unfiltered) and dissolved (filtered) fraction. Samples are collected for the analysis of trace elements, trace organics and toxicity. In addition, ship-board measurements of general water quality are taken, including conductivity/temperature/depth (CTD) profiling of the water column.

The University of California, Santa Cruz (UCSC) performs analysis of total and dissolved trace elements and ship-board water quality measurements. The University of Utah (UU) performs analysis of total and dissolved organics. In addition, Pacific Eco-Risk Laboratories (PERL) performs toxicity analysis. Except for preparation of samples and ship-board measurements of water quality parameters, all samples are processed in the laboratory after the cruise.

The objectives of the water cruise are:

- Collect water samples from 26 sites for analysis of total and dissolved trace elements.
- Collect water samples from 18 sites for analysis of particulate and dissolved organic contaminants.
- Collect water samples from 26 sites for analysis of salinity, total suspended solids, chlorophyll a, nutrients (ammonium, nitrate, nitrite, orthophosphate, silicate), and dissolved organic carbon.
- Collect water samples for analysis of total hardness at sites where salinity is greater than 5.0 parts per thousand.
- Collect profiles of water-column temperature, conductivity, salinity, dissolved oxygen, and optical back-scatterance at 24 sites (not done at watershed sites).
- Collect samples for toxicity analysis from six stations.

A minimum of six crew members (excluding vessel captain) are required to conduct a water cruise, although seven crew members are recommended. Cruise member responsibilities are presented in Table 7.

**Table 7. Crew responsibilities for RMP water sampling cruise.**

<i>Cruise Members/Number of Crew (Contractor)</i>	<i>Responsibilities</i>
Applied Marine Sciences/1 (AMS)	Cruise management, collection of As, Hg, Se samples, CTD operation and verification of sample record logs
University of California Santa Cruz/2-3 (UCSC)	Trace element sampling, cognate sampling
University of Utah/2 (UU)	Trace organics sampling
University of California Santa Cruz/1 (UCSC)	Vessel operation

The Prime Contractor (AMS) is responsible for oversight of sampling operations, compliance with cruise plan and quality assurance guidelines, maintenance of the sample field log, preparation of chain-of-custody records, and operation of the CTD. UCSC is responsible for all trace element sampling and collection of cognate samples. UU is responsible for collection of all trace organic samples. In addition, UCSC provides the vessel and skipper.

### **3.1.2 Water Sampling Vessel Safety**

The cruise manager develops the cruise plan in coordination with the vessel captain to ensure vessel availability and that tides are appropriate to the work to be conducted. The actual cruise schedule may vary, primarily due to inclement weather. Other factors such as personnel availability and vessel malfunction occasionally require changes to the cruise schedule. Once underway, the skipper and cruise manager consult the cruise plan and incorporate changes induced by weather or other factors to determine which sites are to be sampled on that day.

The vessel skipper is responsible for navigating the vessel to the sample collection sites. Once at a sampling site, a crewmember (usually the cruise manager) will be instructed by the vessel skipper to deploy the anchor. The vessel will swing into the current within a few minutes, at which time the cruise manager will record the vessel position and time of arrival in the cruise logbook. The vessel coordinates are checked against the coordinates listed in the cruise plan.

The vessel captain is responsible for overseeing the safety of the vessel and crew while they are onboard the vessel. It is the responsibility of each crewmember to follow common safety practices while performing their duties. Safety practices include but are not limited to:

1. Participants on the rear deck (outside the vessel cabin) are required to wear a Personal Flotation Device (PFD).
2. Participants using hazardous chemicals (i.e. acid or methanol) are required to wear appropriate Personal Protection Equipment (PPE) such as gloves and eye protection.
3. In locations where contact of the skin to the sample water may pose a health risk, participants are required to wear appropriate gloves and thoroughly wash after contact with estuary water.
4. Participants are required to store equipment and personal belongings in a safe manner.
5. Participants susceptible to motion sickness are advised to take appropriate steps to minimize its effects.
6. Tripping hazards must be minimized by routing electrical cords and sample lines away from areas of high foot traffic.
7. Participants are required to notify the vessel captain and cruise manager of any vessel, operational or personal safety concerns.

### 3.1.3 Water Sampling Equipment List

It is the responsibility of each participating laboratory to prepare their equipment prior to the sampling cruise. Normally, trace element sampling equipment is prepared by UCSC, trace organics sampling equipment is prepared by UU and miscellaneous support equipment is prepared by AMS. The equipment list for water trace elements sampling is provided in Table 8, water trace organics sampling is provided in Table 9, and the miscellaneous support equipment for water toxicity sampling is provided in Table 10.

**Table 8. Equipment list for water trace elements sampling.**

<i>Quantity</i>	<i>Description</i>
<b>General Equipment</b>	
1	15-20 foot sampling pole (2 pieces; 1 hollow, 1 with a telescopic insert)
1	Solomat™ (for field measurements of salinity, conductivity, pH, dissolved oxygen, and temperature)
2	Sampling buckets
2	Chairs
25	Filter cartridges
2	Ice chests containing: 20 pounds dry ice/sampling day
1	Cruise log book and cruise plan
1	Tape: (2) plastic, (6) labeling
1	Pens: (6) ballpoint, (6) Sharpies
1	Razor blades
1	Trash bags (recycled from lab)
1	Ziploc™ bags (50 each size; 9 x 12, 12 x 15)
1	Nylon gloves (4 pair, medium, 2 pair, small)
1	Polyethylene gloves (6 boxes, medium, 3 boxes, small)
6	Kimwipes™ and K-dries™, large (3 boxes each)
1	Sun-protection lotion
1	Spare batteries
1	Bucket opener
<b>Water Sampling System</b>	
1	Masterflex™ dual-head peristaltic pump
2	Pump speed controller boxes
4	Pump heads

<i>Quantity</i>	<i>Description</i>
1	Spare pump head screws
1	Spare pump fuses (3 amp)
2-3	Electrical power extension cords, 25 foot
1	Flexframe™ fittings and clamps (2 small to hold tubing and 2 large for filter cartridges)
1	1/2 inch aluminum support rod and plastic covered base
1	Tape: plastic, duct, electrical
1	Hand tools: screwdriver, pliers, crescent wrench
3	Aluminum adjustable (screw-tighten) hose clamps (to hold sample poles together)
as needed	Straps, bungee cords, nylon rope
3 sets	Pre-assembled acid-clean tubing, consisting of the following: <ul style="list-style-type: none"> <li>• Inlet tubing: 25 feet of 5/16 inch ID Teflon™ tubing</li> <li>• Outlet tubing: 3 feet of 5/16 inch ID Teflon™ tubing</li> <li>• Pump-head tubing: 2 pieces, 1 foot long of 5/16" ID C-flex tubing</li> <li>• 2 - "Y" polypropylene fittings (to connect 5/16" ID tubing)</li> </ul>
1	Ziploc™ tool
as needed	Cable ties
1	Gloves: nylon (inner), polyethylene (outer)
	<b>Chlorophyll Filtration System</b>
2 sets	Filtration flask with vacuum tubing
2 sets	Filter holder (reservoir, stoppered neck, clamp)
60	Glass fiber filters (47 mm)
1	Graduated cylinder (100 ml)
2	Forceps, filter-type
1	Rinse bottle
1	Glass-distilled water (2 – 2 L) (refill)
2 boxes	Centrifuge tubes, 15 ml screw-cap polycarbonate
50	Ziploc™ plastic bags (4 x 6), to hold 2 tubes per site
1	Tube rack
1	Aluminum foil
1	Hand-vacuum pump
	<b>Chromium Sampling Equipment</b>
60	Dissolved Cr sample bottles, 140 ml low density polyethylene (LDPE), filled with 1% HCl
1	Styrofoam blocks and bungee cords to support filtration setup
1	1 L LDPE sample collection bottle for particulate Cr
2	Vacuum filtration flasks with vacuum tubing
2 and spare	Filter flasks, stoppered
1	Eppendorf™ pipette, 1,000 ml with large tips
1	Graduated cylinder, 250 ml plastic
2	6N HCl (140 ml LDPE bottles)
1	Safety goggles
1	Sodium bicarbonate in rinse bottle
1	Ultrapure water (2L), prepared in laboratory
1	Rinse bottle (for Milli-Q)
60	Nucleopore™ filters, acid-cleaned and each preloaded in 4 ml LDPE bottles
1	O-rings (stored in weak HCl in 60 ml jar)
2	Nylon tweezers (stored in weak HCl)
7	Dissolved Cr travel blanks, ultrapure water in 140 ml LDPE bottles
1	Polyethylene gloves
1	Chromium log book
1	Hand vacuum pump

**Table 9. Equipment list for water trace organics sampling.**

<i>Quantity</i>	<i>Description</i>
1	Axys™ organics sampler (custom manufactured for total and particulate fraction sampling)
1	Cruise log book
1	Drip pan
1	Sampler repair kit
1	Sampler manuals
1	Cotton tipped swabs
1	Micro-pump service repair kit
1	Extra screws, seals, and bushings
1	Bushing extractor and inserter
1	Magnet set height tool
1	Precision forceps
1	Torque wrench
1	Silicone lubricant
1	Sample tubing
1	Intake/Exit tubing
1	Spare intake tubing
1	Spare solvent rinsed tubing
1	Columns loaded with XAD-2 resin
1	Glass fiber filters, kilned and rinsed
2	Coolers with column racks
2	Coolers for dry ice
2	Pack sample gloves and dry ice gloves
1	Plastic work box
1	Column assemblies closed with 2 tube to tube Gorilla-Grip™ unions
2	Jars each with 4 tube to column Gorilla-Grip™ unions
1	Plastic waste beaker
1	Container of spare Swageloc™ parts
1	Container of pre-filters, 140m filter cups, and spare Gorilla-Grip™ unions and ferrules
1	Safety glasses
1	Heavy duty aluminum foil
2	Solvent boxes
1	Large Kay Dry™ wipers
1	Plastic bags for filters
as needed	Cable ties
as needed	Garbage bags
1	Solvent squirt bottles for methanol
1	Bag of pens, sharpies, time tape, Teflon tape, and water proof hockey tape
1	Mobile tool box with tools
2	Wooden column racks
2	Empty calibrated carboys with lids and handle
5	Carboys of distilled or reverse osmosis drinking water
6	Bottles of methanol (4L)
2	Towels
1	Rope
1	Extension cord

**Table 10. Miscellaneous support equipment list for water toxicity sampling.**

<i>Quantity</i>	<i>Description</i>
1	SBE1 19 CTD, calibrated in the laboratory prior to use
1	Data terminal and communication cable for CTD
1	CTD Maintenance kit including 8 new “D” size batteries
3	Igloo™ coolers
8	Pre-cleaned, 10 L polyethylene carboys; fluoride coated
1	Extension cord
1	Extendable aluminum pole
1	Pack of Nalgene™ tubing, factory cleaned
20	Lbs. Ice
1	110 VAC Masterflex™ peristaltic pump
1	Duct tape

### 3.1.4 Sample Containers

The containers required for water sampling are listed in Table 11.

**Table 11. Container list for water sampling.**

<i>Sample Type</i>	<i>Container</i>
	<b>Trace Elements Sampling</b>
Trace metals	2 per station: (56) 1 L and (4) 2 L LDPE (packed in eight 5-gallon buckets)
Nutrients	3 per station: (75-100) 125 ml LDPE (packed in one 5-gallon bucket)
Total suspended solids	(30) 500 ml, 1 L or 2 L LDPE (packed in one plastic box or ice chest)
Salinity	(30) 500 ml LDPE (packed in one plastic box or ice chest)
Dissolved organic carbon	2 per station: (60) 30 ml glass vials (packed in ice chest along with small Styrofoam box)
Chlorophyll	2 per station: (60) 15 ml plastic centrifuge tubes (packed in chlorophyll equipment box)
	<b>Trace Organics Sampling</b>
Total organics	Pre-cleaned columns (2 per site)
Particulate organics	Pre-cleaned filters (number used per site depends on turbidity, normally 1 per site)

### 3.1.5 Trace Elements Sampling Equipment Preparation

The equipment used for water trace elements sampling consists of a peristaltic sample pump, Teflon™, C-flex™ and polypropylene sample tubing and fittings, trace metals filter (for dissolved trace metals sampling) and assorted sample containers. The following sections outline the preparation for the trace metals sampling system components and sample containers.

#### Teflon™ Sample Tubing

The main intake tubing for trace elements sampling is composed of Teflon™ tubing. This tubing is cleaned prior to use at each sample location. A Masterflex™ peristaltic pump is used to fill the Teflon™ sample tubing with reagents and to flush with water between acid cleaning. The Teflon™ tubing ends are joined together with C-flex™ tubing to hold reagents for soaking

overnight or between sample sites. Advance preparation of the Teflon™ sample tubing is done in the laboratory 3-4 weeks before the start of a cruise and is also done between each sample site. The following procedures are used for cleaning Teflon™ sample tubing:

- Completely fill tubing with laboratory-prepared Micro solution and let soak 3-7 days at room temperature (60-70 °F).
- Drain Micro solution from the tubing and flush with approximately 10 liters (L) of deionized water (DI).
- Completely fill sample tubing with 6 normal (N) reagent-grade hydrochloric acid (HCl) and let soak 7 days at room temperature.
- Drain acid and flush tubing with ultrapure water until pH of the effluent is about 5.0 units to ensure that HCl is completely flushed out.
- Completely fill tubing with 7.5 N reagent grade nitric acid (HNO<sub>3</sub>) and let soak 14 days at room temperature.
- Drain acid and flush tubing with ultrapure water until pH is approximately 5.0 units.
- Fill tubing with ultrapure water that has been acidified to pH 1.0 with trace metal grade HNO<sub>3</sub> or HCl. Cap the ends of the tubing with C-flex™ and store tubing filled with this solution. Keep solution in tubing until used in the field.

Between each sample site and at the end of the sampling day, the Teflon™ sample tubing is thoroughly cleaned and stored using the following procedures:

- Empty the sample tubing of any residual sample water by pumping with air and flushing with ultrapure water.
- Flush tubing with 1 L of reagent grade methanol. Fill completely with methanol and let set 15-20 minutes. Drain the methanol from the sample tubing, flush with air then flush with 2 L of ultrapure water.
- Completely fill tubing with 3 N trace metal grade HCl and let sit 1-2 hours (this should be accomplished during the transit between sample sites). Drain acid from the sample tubing and flush with several liters of ultrapure water. For long-term storage (overnight during sample cruise), store tubing filled with weak acid solution.
- Cover the ends of the tubing with 2 clean polyethylene gloves. Place tubing in a cleaned Ziploc™ bag and label as “clean”.

## **C-Flex™ Pump Tubing**

Preparation is done in the laboratory, four weeks before the start of a cruise. The following procedures are used for cleaning C-flex™ pump tubing:

1. Cut C-flex™ tubing into 6" and 1' lengths.
2. Soak in laboratory-prepared Micro solution for 24 hours.
3. Rinse tubing with DI water. Soak tubing in hot 3 N reagent grade HCl for 24 hours.
4. Rinse tubing with ultrapure water. Soak in hot 4 N reagent grade HNO<sub>3</sub> for 24 hours.
5. Rinse tubing with ultrapure water. Check to ensure that tubing has not lost its integrity.

## **Polypropylene Sample Tube Fittings**

Preparation is done in the laboratory, four weeks before the start of a cruise. The following procedures are used for cleaning sample tube fittings:

1. Soak fittings in laboratory-prepared, hot Micro solution for 24 hours, or at room temperature for seven days.
2. Rinse fittings with DI water and soak in 6 N reagent grade HCl for seven days.
3. Store fittings in weak trace metal grade HCl for 1-2 weeks or until used.
4. Rinse fittings with ultrapure water prior to use.

## **Trace Metal Filter Cartridges**

Filter cartridges are used for collection of dissolved trace metal samples in water. Normally, one filter cartridge is used per sample site, although elevated suspended solids concentrations may require using two or more cartridges per site. Filter cartridges are prepared in the laboratory, brought into the field, exposed to the sample medium, and taken back to the laboratory for sample extraction and analysis. Preparation of filter cartridges is done two weeks before the start of a cruise. The following procedures are used for cleaning filter cartridges:

1. Cut at least 50, 4-5" sections of Micro-cleaned C-flex™ tubing. Two pieces of tubing are required for each filter cartridge used.
2. Cut at least 25, 1.5" pieces of acid-cleaned Teflon™ tubing (5/16" ID). One piece will be needed per cartridge.
3. Connect C-flex™ tubing sections to each side of the filter cartridge. Connect Teflon™ tubing to one side of the filter cartridge/C-flex™ tubing assembly. Repeat steps for all filter cartridges.
4. Connect several filter cartridge/tubing assemblies together in a chain with the red flow arrows all pointing in the direction of the flow.
5. Wet the cartridges with reagent grade methanol. Using a Masterflex™ peristaltic pump, fill cartridges with reagent grade methanol and let sit for several minutes.
6. Pump out methanol by first pumping system with air, then flush thoroughly with 4 L of ultrapure water. Flush with air, then 4 L of ultrapure water, then air again. Fill cartridges with ultrapure water and let sit for several minutes, then drain completely. It is necessary that no

trace of solvent remains as traces of organic solvent mixed with acid in a closed bottle can become explosive.

7. Fill cartridges with 3 N reagent grade HCl and let sit 7 days, at room temperature.
8. Pump out acid. Flush with approximately 10 L of ultrapure water, until the pH is about 4.0.
9. Fill cartridges with 4 N trace metal grade nitric acid and let sit 7 days.
10. Pump out acid. Flush with approximately 10 L of ultrapure water, until pH is about 4.0. Store cartridges filled with this weak acid solution inside until use. Separate cartridges during this step, and continue flushing individually, to ensure that each cartridge is stored with a weakly acidic solution.
11. Separate cartridges so that there is a piece of C-flex™ tubing on both the inlet and outlet. By looping the C-flex™ tubing downward (consider flow direction as indicated by red arrow, and have the vent in an upward position) connect the two pieces with the piece of Teflon™ tubing between them. Rinse off outside of looped assembly with de-ionized water.
12. Bag acid-cleaned cartridges individually in Ziploc™ polyethylene bags and store in a clean 5 gallon plastic bucket.

### **Trace Metal Clean Sample Bottles**

Preparation is done in the laboratory, 4-8 weeks before the start of a cruise. The following procedures are used for cleaning sample bottles and labware:

1. With a diamond-tip stainless steel scribe, etch the cap, side and shoulder of each sample bottle with a unique identification number. One and two liter bottles have separate numbering systems. Record numbers in field log book.
2. Place bottles in a bath filled with Micro solution (completely immerse with no air bubbles). Keep immersed for 2-7 days at room temperature.
3. Rinse bottles 5-6 times using approximately 10% of bottle volume with tap water, rinsing bottle exterior at same time, including cap and bottle threads. Make sure the final rinses no longer foam when shaken. While rinsing, check seal of cap (toss leaky ones).
4. Rinse bottles 2-3 times with DI or ultrapure water and store in large un-pigmented polyethylene bags until next procedure.
5. Wipe down fume hood and set inside a large plastic secondary containment tray. The tray should be pre-rinsed with ultrapure water and is stored in a large polyethylene plastic bag when not in use. Set Micro-clean bottles in the tray. Fill bottles to the brim with 6 N reagent grade HCl. Tighten caps excluding any air bubbles. Tilt bottle at approximately a 30 degree angle and squeeze sides while screwing down cap. Rinse outside of capped bottle with ultrapure water. The tray catches any spilled acid and water.
6. Place acid-filled bottles into a 2 N HCl bath for at least 2 weeks. The 2 N HCl baths are made with reagent grade HCl and DI water. Acid bath containers are 5 gallon white plastic buckets cleaned with micro and deionized water. Three 2 L or six 1 L bottles fit in one 5 gallon acid bath.
7. In a clean plastic containment tray set in the fume hood, place a batch of Micro-clean bottles (three 2 liter or six 1 liter). Remove a batch of acid-cleaned bottles from 2 N HCl, rinsing the exterior with ultrapure water over a plastic rinse tray and place in fume hood next to Micro-clean batch. Pour the 6 N HCl from these bottles into the next batch of bottles to be acid-cleaned. Rinse cleaned bottles with ultrapure water to remove most of the acid and store in

large polyethylene bags. The 6 N reagent grade HCl can be re-used at most 6-7 times, as long as bottles are pre-rinsed well enough to remove micro residue. If the acid foams at all, discard the acid.

8. In a clean lab, rinse exterior of bottles with ultrapure water before opening. Rinse the inside of bottles with ultrapure water 5 times (using about 10% of bottle volume each rinse). Drain each rinse into the cap then pour this over the threads of the bottle.

For sample bottles used at the Golden Gate sampling site or other “low level concentration” oceanic sampling sites:

1. In the clean lab, fill bottles with 7.5 N HNO<sub>3</sub> and let sit for one week. Rinse with ultrapure water 5 times.
2. Fill each bottle to the base of the neck with ultrapure water and place in a class 100 (HEPA) work area. Add approximately 10 ml of concentrated trace metal grade HNO<sub>3</sub> per each 1 L bottle, then fill to brim with ultrapure water. Tighten cap excluding air bubbles. Make sure cap is secure. Invert to mix.
3. Double bag each bottle in polyethylene Ziploc™ bags. Wipe down interior bags with Kay-dry™ to remove any dust particles. Write bottle identification number on interior bag.
4. Store four 2L or eight 1L bottles in two large polyethylene bags inside 5 gal polyethylene buckets.
5. Bottles are set for 2 to 3 weeks filled with weak HNO<sub>3</sub> prior to use.

### **Recycling Trace-Metal Sample Bottles**

Trace metal sample bottles may be recycled after use. The laboratory is responsible for determining when sample bottles may be recycled and when they should be discarded.

Preparation of bottles for recycling is done in the laboratory 2-3 weeks before the start of a cruise. Use the following guidelines for recycling trace metal sample bottles:

1. Conduct entire cleaning process in class 100 (HEPA) work area and wear polyethylene gloves throughout the procedure.
2. Remove old labels with methanol.
3. Rinse bottle exterior with ultrapure water before opening. Empty contents from all sample bottles. Rinse one bottle with ultrapure water 3-5 times.
4. Fill this sample bottle with freshly made trace metal grade 6 N HCl. Fill the bottle half-full with ultrapure water, then fill remaining half (to neck) with concentrated trace metal grade HCl (12 N) to make a 6 N solution. Shake and swirl bottle so acid touches all inside surfaces, then set bottle upright.
5. While acid is sitting in this bottle, rinse another bottle with ultrapure water 3-5 times.
6. Decant the 6 N HCl from the first bottle into the bottle just rinsed with ultrapure water. Cap and re-bag bottle just cleaned with HCl. Shake and swirl HCl in second bottle then set aside while the next bottle is rinsed with ultrapure water.
7. Continue to rinse all bottles with same batch of 6 N HCl. Rinse outsides of bottles with ultrapure water before opening.

8. Rinse inside of bottles with ultrapure water 3-5 times (using 10% of bottle volume each rinse). Drain each rinse into the cap then pour this over the threads of the bottle. Rinse the bottle exterior also.
9. Fill each bottle to the base of the neck with ultrapure water. Add approximately 10 ml of concentrated trace metal grade  $\text{HNO}_3$  for each liter bottle.
10. Tighten cap, careful to exclude air bubbles. Make sure cap is secure.
11. Double bag each bottle in Ziploc™ bags. Wipe down interior bags with Kay-dry™ to remove any dust particles. Write bottle identification number on interior bag.
12. Store four 2 L or eight 1 L bottles in two large polyethylene bags inside 5 gal polyethylene buckets or in plastic ice chests. Bottles are set at least 2-3 weeks before use.

### **Teflon™ Bottles for Hg, As, Se Samples**

Preparation is done in the laboratory 4-8 weeks prior to the start of a cruise. Use the following guidelines for cleaning Teflon™ bottles:

1. Number each bottle with a diamond-tip stainless steel scribe. Etch the cap, side and shoulder of each bottle with a unique identification number. One liter and two liter bottles have separate numbering system. Record numbers used in field log book.
2. Place bottles in micro bath for at least 2-7 days at room temperature. Rinse inside and outside of bottle 5-6 times using approximately 10% of bottle volume with DI water, including cap and bottle threads. Make sure final rinses no longer foam when shaken. While rinsing, check seal of cap and discard leaky ones.
3. Rinse with ultrapure water 3-4 times and store in large, unpigmented polyethylene bags until ready for next step.
4. In a fume hood with hood fan on, fill bottles to brim with concentrated reagent grade  $\text{HNO}_3$ . Tighten caps excluding any air bubbles. Tilt bottle at approximately a 30 degree angle and squeeze sides while screwing down cap, do this over clean rinse tray.
5. Place acid-filled bottles into a 7.5 N reagent grade  $\text{HNO}_3$  bath and let sit 4 weeks at room temperature or 1 week at hot temperature (60-80 °C). For a hot acid bath, place a 4 L glass beaker on hot plate in fume hood.
6. In a fume hood, place a batch of micro-clean bottles. Remove a batch of acid-cleaned bottles from 7.5 N  $\text{HNO}_3$ , rinsing the exterior with ultrapure water over a plastic rinse tray and place in fume hood next to micro-clean batch. Pour the concentrated  $\text{HNO}_3$  from these bottles into the next batch of bottles to be acid-cleaned. Rinse cleaned bottles with ultrapure water to remove most of the acid and store in large polyethylene bags.
7. In a clean lab, rinse exterior of bottles with ultrapure water before opening. Rinse the inside of bottles with ultrapure water 5 times (using about 10% of bottle volume each rinse). Drain each rinse into the cap then pour this over the threads of the bottle.
8. Fill each bottle to the base of the neck with ultrapure water and place in a class 100 work area. Add approximately 15 ml of concentrated trace metal grade HCl per each 1 L bottle (fill to brim with ultrapure water). Tighten cap excluding air bubbles. Make sure cap is secure.
9. Double bag each bottle in polyethylene Ziploc™ bags. Wipe down interior bags with Kay wipes™ (moistened with ultrapure water) to remove any dust particles. Write bottle identification number on interior bag.

10. Store four 2 L or eight 1 L bottles in two large polyethylene bags placed inside a 5 gallon polyethylene bucket. Bottles will set 2 to 3 weeks filled with weak HNO<sub>3</sub> prior to use.

In addition to the above sample containers, bottles are used for collection of ancillary samples such as total suspended solids, salinity and chlorophyll. Ancillary sample bottles normally are prepared in the laboratory 4-8 weeks before the start of a cruise. Ancillary sample bottles are rinsed with deionized water and air-dried. Ancillary sample bottles may be reused.

### **3.1.6 Trace Organics Sampling Equipment Preparation**

The equipment used for water trace organics sampling consists of an Axys™ organics sampler (custom manufactured to sample both particulate and dissolved fractions at the same time), XAD-2 resin columns (filtered and unfiltered) for organics extraction, and a sample tubing system composed of Teflon™ tubing and Swageloc™ stainless steel fittings. Other than the columns used for sampling total organics and the filters used for sampling particulates, no containers are used for sample collection. Resin for filling Teflon™ columns and filters for sampling particulates are prepared in the laboratory at the University of Utah at least four weeks before the start of a cruise.

Current sampling protocol requires three batches of resin to be prepared for each cruise. These resin batches remain discrete throughout sampling and extraction. Each batch of resin fills 16-17 sample columns. Normally, one batch of resin is used to sample the “clean” central bay sites, another to sample the “dirtier” north bay sites (not including Petaluma River), and the third for the “dirtiest” south bay (plus Petaluma River) sites. When the sample columns are extracted, each set of columns becomes one extraction set with one set of norms and two extraction blanks, always from the same batch of resin. This minimizes the number of norms, and minimizes variation within the set and between samples and blanks. Extracting the sets in order from cleanest to dirtiest also minimizes potential cross-contamination between sets.

### **3.1.7 CTD Preparation**

Pre-cruise preparation and calibration of the CTD takes place at least three days prior to the start of a cruise. Refer to the SeaBird™ SBE-19 CTD operators manual for a full description of calibration and maintenance procedures. Use the following guidelines for inspecting the CTD:

- Visually inspect the CTD for abnormal wear or corrosion. Check to ensure that the deployment line is secure and that there are no abrasions. Make sure all fittings are secure and all fasteners are firmly in place.
- Verify that the communication connector is secure, waterproof and lubricated with silicone grease.
- Verify that the CTD communicates correctly with the data terminal by running the terminal emulation program (TERM19).
- Check the CTD battery status and replace main batteries if voltage is below 6.0 volts.
- Calibrate the dissolved oxygen (DO) sensor.

Calibration of the DO sensor requires measuring the oxygen current output in a zero oxygen environment (sensor purged with nitrogen gas) and in an air-saturated environment (sensor in a 100% oxygen saturated bath). Calibration of the DO sensor requires the following steps:

1. Fill a calibration bath (55-gallon plastic trash can) with tap water and aerate the bath with an airstone placed no greater than 10 cm from the surface. Aerate the bath for at least 24 hours prior to calibrating the CTD. Moderate aeration is maintained to avoid air supersaturation. The water temperature of the calibration bath is kept between 19-21 °C.
2. Measure the zero point oxygen voltage by flushing the sensor with a continuous stream of nitrogen gas. Insure that power has been applied to the sensor for several minutes before the gas is placed in the sensor. Connect the CTD to the data terminal and run the SEASAVE software program to display real-time oxygen current data. Watch the output of the sensor decrease rapidly towards zero volts. Record the voltage output after three minutes. This will be the zero value to use in the calibration. The original calibration sheet that accompanied the oxygen sensor will contain the zero oxygen current that was obtained during the factory calibration. You may compare the results of the recent calibration to those obtained at the last factory calibration.
3. Measure the air-saturated oxygen voltage by immersing the CTD into the calibration bath. Leave the CTD in the calibration bath for at least one hour and do not turn the power on to the CTD. After one hour, turn on the CTD power for at least 12 minutes and record the oxygen current output using the SEASAVE program. It is important to make sure that the CTD pump is working and water is being pumped across the DO sensor membrane. Make sure the CTD remains fully immersed during the calibration procedure. Turn off the CTD when the oxygen current is recorded.
4. OXFIT is the software program used to compute new oxygen sensor calibration coefficients. Run the OXFIT program and fill in the following information:
  - Local barometric pressure obtained from the Livermore Airport (or nearest facility), not corrected to sea level.
  - Water temperature of the calibration bath.
  - Oxygen current in the air saturated water (calibration bath).
  - Oxygen current at the zero point oxygen level.
5. The OXFIT program will calculate a new oxygen current bias and slope that are used to compute the actual oxygen concentration. Refer to the SeaBird SBE19 owners manual for details on the algorithm used to compute actual oxygen concentration. The new bias and slope coefficients are compared to the original factory calibration or the last calibration that was performed. Typically, slope values will slowly increase with time as the sensor is used. The new bias and slope values are entered into the SEASOFT.CON file using the SEACON program.

During normal sampling on the Estuary, care must be taken to avoid fouling the CTD oxygen membrane with organic residue. The oxygen sensor may be rinsed with a 1 % solution of Triton X™ (kept in the CTD field kit) and flushed with deionized water at the end of each cruise day. Store the CTD overnight by filling a plastic syringe with deionized water and injecting the

water into the CTD intake port. Make sure that the DO sensor is immersed in deionized water for overnight storage. For routine cleaning, soak the oxygen sensor in a 1% solution of Triton X™ that has been warmed to a temperature of 50 °C. After soaking, drain and flush the CTD with warm fresh water for one minute. Always store the CTD with the dissolved oxygen sensor fully immersed in deionized water.

### **3.1.8 Water Sampling Procedures**

#### **3.1.8.1 Water Trace Metals Sampling**

Water trace metals sampling is conducted after the vessel anchors in the correct position for the sample site and the captain switches off the engines. Sampling requires set-up of a Flexframe™ support rod and base which is assembled and secured onto the starboard ledge at the rear deck of the vessel. The Flexframe™ assembly is installed prior to sampling at the first site of the day and left in place for the day. All trace metals sampling equipment is brought inside the vessel cabin for overnight storage. A dual-head Masterflex™ peristaltic pump is used to pump sample water from the Estuary into the sample bottles. The pump is assembled with the C-flex™ tubing portions of sample tubing passing through the pump heads. The pump and a pump speed controller are secured just below the Flexframe™ assembly and are connected to the vessels power source with an extension cord.

An aluminum sampling pole is assembled and the sample tubing inlet is secured to one end, with the tubing tip hanging loose approximately two feet off pole end. The sample tubing outlet is secured to the Flexframe™. During sampling, a plastic cover is removed from the inlet end of the Teflon™ sample tubing and the sampling pole is extended over the windward side of the vessel. The pole is oriented up-current from the vessel and upwind from all equipment and personnel. The sample tubing inlet is submerged approximately 1-2 feet into the water column. Surface water is pumped through the sample tubing and the entire tubing system is flushed continuously for approximately 5 minutes before any samples are collected. Both filtered and unfiltered water is collected at each site.

Two persons are needed to conduct the sampling. The “dirty hands” person assists the primary “clean-hands” sampler by controlling the flow controller for the peristaltic pump, holding on to or adjusting the sample pole, adjusting the outlet tubing or filter cartridge, and handing sample containers to the “clean hands” person. The “dirty hands” person does not touch the trace-metal clean bottles, but opens the Ziploc™ bags so that the “clean hands” person may remove them from the bags. The “clean hands” person, wearing at least one pair of polyethylene gloves, does not touch anything with her/his hands except the inner Ziploc™ bag, the bottles and the water.

The clean hands/dirty hands system is not critical for the ancillary samples, and these bottles may be rinsed just three times with sample water before collecting the sample. For trace element samples, the “clean hands” person drains the ultrapure water from the pre-cleaned sample bottle onto the bottle cap and pours the remaining ultrapure water over the bottle threads several times. The sample container is rinsed with the sample water five times, then filled up to

the “neck” with sample water. The sample collection sequence for total trace metals (unfiltered) sampling is as follows:

- Total suspended solids (TSS)
- Chlorophyll
- Total chromium
- Total bulk metals
- Total As, Hg, Se

After the above samples are collected, the filter cartridge is attached to the sample tubing outlet and secured to the Flexframe™. The storage solution inside the filter is drained and the entire sample tubing and filter assembly is flushed with sample water for five minutes. The sample collection sequence for dissolved trace metals (filtered) sampling is as follows:

- Salinity
- Dissolved chromium
- Dissolved bulk metals
- Dissolved As, Hg, Se
- Dissolved organic carbon

A filtered water sample is collected in duplicate to evaluate precision of the sampling equipment and to assess short-term environmental variability at the sample site. Ten percent of all RMP sample stations are collected in duplicate.

### **3.1.8.2 General Water Quality Measurements**

General water quality is measured using a Solomat™ 520C multi-functional chemistry and water quality monitor. This hand-held monitor has several probes which are submerged approximately 3 feet into the water column to collect readings. A multi-parameter probe measures water temperature, depth and conductivity. A dissolved oxygen probe measures dissolved oxygen and a pH probe measures pH. The meter is calibrated for conductivity with a KCl standard, dissolved oxygen using a mixture of  $\text{CoC}_{12}$  and  $\text{NaSO}_3$  and for pH using buffers of pH 7 and 10.

### **3.1.9 Water Organics Sampling**

The Axys™ organics sampler is cleaned before use at each site. To clean the sampler, the sample intake line is placed into a 20 L carboy filled with DI water. The sample line is flushed with enough DI water to rinse the lines and each filter housing with approximately 3 L of water. Sample lines are then drained by removing the intake line from the carboy and running the sample pump dry. The filter cups are removed from the sampler and the water is dumped from the cups. With the filter cups replaced, the sampler is rinsed with 2 L of ACS grade methanol drawn through the intake line using the sample pump. With both the inlet and outlet of the sample line placed in the methanol bottle, methanol is cycled through the filter housing for a minimum of three minutes. The sample lines are drained and the filter cups again are emptied. Each filter

cup is rinsed three times with methanol to remove any sediment before it is replaced. The sampler and both filter cups are rinsed with another 2 L of distilled water. The sample lines and filter cups are drained of water. Sample lines, filter cups and “O” rings are rinsed three times with methanol then replaced. A clean filter is placed in each filter cup, the columns are attached and the sampler is ready to sample.

After the sampler has been used at the first site of the sampling day, there is still a clean filter in one housing, with the other housing containing the collected particulate sample. The filter is removed, wrapped in foil, labeled, bagged, and placed on dry ice. The filter cup is rinsed with methanol to remove any sediment and replaced empty. The column assembly is removed and the columns are capped, taped, and placed in a chilled cooler. The sampler and the empty filter cup is flushed with bay water at the next sampling site with approximately 2 L of water to avoid sample contamination. The column assembly with a clean set of columns is attached, the filter valve is switched to the clean filter and the next sample is collected.

If the flow rate is slowed and the water or the changed filter are not particulate laden, then a pre-filter clog is indicated. To remove and clean the pre-filter, the sampler is turned off and the inline sample line and pre-filter are removed. The pre-filter is taken apart and the parts are rinsed with methanol. The pre-filter cup is replaced if it has holes or looks compromised. The pre-filter is reassembled in the order in which it came apart, the cup must be seated properly or the pre-filter will leak.

The sampler is placed on the starboard bench inside the boat’s cabin and secured with straps. The exit tube and the intake tube are attached to the proper Swageloc™ ports on the front panel of the sampler. The intake tube has a (pre-cleaned) stainless steel mesh screen on the intake end and an inline pre-filter with a directional arrow situated appropriately on the other end between the intake and the intake port.

When the vessel is at anchor with the engines off, the cabin doors and windows can be open while assembling the columns and inserting the filters. When the vessels engines are running, the cabin doors and windows must be closed before opening any internal part of the sampler, to reduce the possibility of airborne contamination (i.e. diesel exhaust). If the sampler has been cleaned, then two clean filters are loaded into the filter cups. A piece of tape, on each filter housing, indicates that a clean filter is loaded. The tape is removed from the housing after the filter has been used. After the first sampling site of the day, one filter cup is left empty so that the rinse water can be routed through the empty filter cup as the sampler is rinsed with water from each site prior to sample collection.

A large plastic box is lined with clean aluminum foil. Each column is labeled with the site code, the date and the collector’s initials, and covered with 2 inches of transparent tape. The column end-caps are removed and placed into a clean jar. The methanol is drained from each column and the 4 female Gorilla-Grip™ connectors are attached to the ends of each column. The straight unions are removed from the column assembly and the ends of the metal tubing are rinsed with methanol and placed in the open ends of the female Gorilla-Grip™ connectors. The Gorilla-Grip™ connectors are first tightened by hand and then tightened with the proper wrenches another 1 turns. The Teflon™ threads will easily strip if over-tightened. The columns

are assembled so that the labels are in the proper orientation with the A column on the left and the B column on the right. The column numbers as well as the filter batch color are recorded on the sample log sheet for each site.

An XAD column and a filter field blank are collected each week that the sampler is used for RMP samples. To collect a field blank, a filter is unwrapped and left exposed while loading the filter cartridges into the filter cups. For XAD columns, both end-caps of a clean column are removed, the methanol is drained and the column is left open while the columns for that site are loaded onto the column assembly. The blanks are labeled with "FLDBK", the date, collector's initials and the sight code. A sample sheet is filled out for these samples and they are logged in upon return to the University of Utah.

When the sampler is ready for sampling (i.e. sampler is clean or site water has been flushed through if necessary) the column assembly is attached to the sampler. The Swageloc™ unions are tightened to finger tightness, and then tightened 1 turn with the proper tools; over tightening the Swageloc™ connectors ruins them, causing them to leak. The Swageloc™ gauge is used to measure the gap at the joint, and the connectors should be tightened until the gauge just fits in the gap. The filter valve is checked to see that it is pointing towards the cup containing the filter to be used.

The sample intake line is attached to the aluminum pole with cable ties or with Velcro™ ties. The pole is extended to its maximum length, placed over the boat's rail and secured with a bungee cord or tie. The inboard end is strapped to a pad eye in the deck. It is adjusted so that the intake line remains submerged. The intake line is placed about 1.5 to 2 feet under the surface of the water. If the wave action is severe enough that the intake line comes out of the water and takes in air, the pole is held by hand to keep the intake line submerged. The stainless steel screen and about 2 feet of the intake line are rinsed with methanol before and after sampling. Between sites, the foil screen is wrapped with a rinsed piece of heavy duty foil. When motoring between sites, the intake line is coiled up, and it and the pole are brought inside the cabin.

The sampler can usually prime itself if the intake is higher than the exit tube. Allowing the sampler to prime itself when the pump is dry puts excessive wear on the gears, so a hand pump is used to pull the water up to the sampler pump before starting the sampler motor. The sampler is primed by attaching the hand pump intake line to the sampler water exit tube, and the water is pulled through the sampler. Once the start button is pressed, the exit line is quickly removed from the hand pump and placed in a known volume carboy to begin sample collection. The motor RPM is adjusted to maintain a fill time of 10-12 minutes for each 20 L carboy. The RPM changes with total suspended solids (TSS) as well as the motor condition. The approximate RPM is determined from the RPM recorded in the notes from the last sampling. While on the *RV David Johnston*, the RPM remains relatively constant. The RPM for the last sampling is used for the first carboy and the RPM is adjusted based on the time recorded to fill the carboy. The start time is documented, and the sampler is checked for water leaks. Air bubbles in the lines also indicate leaks, and, when detected, can sometimes be stopped by tightening the connections.

If the sampler has a leak that cannot be stopped, this information is recorded in the sample log book. The start and stop time for filling each carboy is documented, and a total of

five carboys are filled at each site. When the fill time exceeds 10-12 minutes, the sampler RPM is increased until the typical fill time is attained. When the water particulate level is high and the pump pressure gauge reads 15 PSI, or higher, the particulate filter needs to be changed. When changing the filter does not improve the flow rate, and the particulate filter looks clean, the pre-filter is inspected and cleaned or replaced as necessary.

After five carboys have been filled, the intake line is removed from the water and the sampler is allowed to run until air bubbles come through the column assembly. The sampler is turned off, and the intake line is rinsed with methanol, covered with foil and coiled onto the pole.

The column assembly is removed from the sampler with the following steps: the columns are removed by loosening the Gorilla-Grips™ with the proper wrenches. Residual water is drained from the columns, and the methanol rinsed end-caps are attached. The end-caps are wrapped with Teflon™ tape and the sample log sheets are filled out with any pertinent information. The columns are placed in a cooler with enough dry ice to keep them cool (usually 5 pounds put in every morning and every evening, depending on ambient conditions).

The filter housing containing the used filter is removed with the following steps:

1. A large pair of forceps is rinsed with methanol and used to remove the filter from the filter housing.
2. The filter is wrapped with a piece of methanol rinsed foil, and is over-wrapped with a second piece of clean foil.
3. The filter is labeled with the collection date, site code and the collector's initials and is then placed in a polyethylene bag which is twisted, wrapped over the filter again, and labeled with the same information on the outside of the bag.
4. The bag is placed in a cooler containing enough dry ice to keep the samples frozen (100 pounds is usually enough to add small amounts to the column coolers and keep the filters frozen for one week of the cruise).
5. On return to the University of Utah laboratory, the filters are placed in a freezer.

The filter cup is re-attached to the sampler without a filter, and is flushed with water from the next site to remove contaminants from the previously sampled site. The filter cup is removed, and the cup and O ring are rinsed three times with methanol. Finally, a clean filter (if needed) is placed in the filter cup, and the filter cup is re-attached to the sampler.

### **3.1.10 Water CTD Profiling**

The objective of water CTD profiling is to measure conductivity, temperature, salinity, optical backscatterance, dissolved oxygen and depth in the water column. A CTD profile is measured from 0.1 meters below the water surface to the sediment layer. CTD profiles are conducted using the following steps:

1. When arriving at a sample site, wait at least five minutes for the vessel to swing into position before turning on the CTD and lowering into the water. The CTD must be initialized with

zero casts prior to use. If casts are held in the CTD memory before deployment, reinitialize the unit before deployment.

2. Turn on the CTD and lower the unit until the top of it is just beneath the surface of the water. If surface waves are greater than one foot, lower the unit three feet below the surface. Record the time the CTD was activated on the sample log.
3. Allow time for the oxygen sensor to polarize and the unit to equilibrate to the ambient water temperature by keeping the CTD just beneath the water's surface for at least three minutes. Failure to wait three minutes may result in erroneously high temperature and oxygen readings.
4. Slowly lower the CTD to the bottom. The CTD's descent rate can not exceed 1.0 meters per second (m/sec) The recommended optimum speed to lower the CTD is 0.25-0.50 m/sec. It is recommended that the scan rate is no less than 1 scan per second and that a descent rate less than 1 m/sec be used. Scan rate is adjusted in the TERM19 program. Optimal scan and descent rates are dependent upon sea surface conditions during deployment and will be evaluated and adjusted accordingly. A cast whose average descent rate exceeds 1 m/sec must be re-sampled.
5. After the CTD reaches the seafloor, it may be brought back to the surface at any speed. Only down cast data are used. Bring the CTD on board the vessel and turn off the power.
6. Connect the communications cable from the data terminal to the CTD. Boot up the data terminal and change the directory to the "C:/SEASOFT" directory within DOS. Run the TERM19 terminal emulation program to communicate with the CTD.
7. Display the CTD status by pressing the "status" function key [F3]. Check that the battery voltage is sufficient (above 6.0 volts) and that the number of casts equals one. Check the CTD header by pressing the function key [F4] and determine that the number of samples is approximately twice the duration of time of deployment (in seconds).
8. Upload the raw data by pressing the "upload" function key [F9]. Each cast receives a unique file name that corresponds to the station code and cruise number. File names can have a maximum of six characters. The program will prompt for header information, which will include the cruise number, station name, station code, vessel name, and operator. Enter this information into the header form.
9. After the raw data has been successfully downloaded, re-initialize the CTD for the next deployment by pressing the "initialize" function key [F8].
10. Copy the raw data file from the computer hard disk to a floppy disk and store the floppy disk in a safe location. Repeat this procedure after every cast.
11. Determine the CTD cast acceptability by running the DATCNV program. If anomalous values are present, the cause must be investigated and remedied before proceeding to the next station. If necessary, resample the site by taking another cast.

### **3.1.11 Water Toxicity Sampling**

Water toxicity samples are collected by using a peristaltic pump with no filter. At the sampling site, factory-cleaned Nalgene™ tubing is fastened to an extendable aluminum pole, allowing 1 foot of tubing to extend beyond the end of the pole. The pole is then placed over the rail of the vessel, allowing the sample tubing to be submerged 1-2 feet below the water. The pole is maintained in a stable position so that the sample tubing remains 1-2 feet below the water column. The outlet end of the sample tubing is placed through the peristaltic pump, leaving

enough tubing on the excurrent side to reach the sample container. The sample container for water toxicity consists of a pre-cleaned, 10 liter polyethylene carboy with the inside surface coated with a fluoride polymer. Sample water is pumped through the tubing for several minutes to flush the tubing, and then a single sample carboy is filled. The carboy is labeled with the site code, date, analysis, and the collector's initials. Water toxicity sample carboys are placed into Igloo™ coolers and held on wet ice until the end of the cruise. Samples are picked up by Pacific Eco-Risk Laboratories (PERL) personnel at the end of the cruise.

### **3.1.12 Watershed Water Sampling**

Watershed water sampling is conducted at two sites located in the southern end of the Estuary. The Standish Dam sampling site is on Coyote Creek, approximately 1.5 miles upstream from its entrance to the bay. The South Bay site is on the Guadalupe River, about two miles upstream from its entrance to the Estuary. Both sites are accessed by car.

The Standish Dam site has no electrical power nearby. Trace metal samples are collected through use of a peristaltic pump with a built in power supply. The organics sampler is provided with 120 VAC power through use of a 12 volt DC deep cycle marine battery coupled to a power inverter. The South Bay site has electrical power, so the inverter is not used there. There are no significant differences in sampling methodologies between sampling from the vessel-accessed sites and sampling from the car-accessed watershed sites. When sampling from the watershed sites, care is practiced to ensure that all work surfaces are kept as clean as possible and sample containers are not exposed to any airborne contaminants during the sampling process.

Watershed sampling typically takes two hours at each site. The longer time is due to greater mobilization times and higher level of suspended solids encountered, which slow the sample pumping volume and require more particulate filter changes during sampling. Sampling the watershed sites typically occurs the day after sampling the last of the vessel-accessed sites. Sampling normally occurs during outgoing tides, though the need to sample both sites on the same day often requires sampling the South Bay site during the start of an incoming tide.

There are minor differences in the set-up of the trace organics sampler for use at the watershed sites. At the Standish Dam site, the water elevation is typically far lower than the elevation of the organics sampler (which is equipped in the back of a van). An extension is added to the sample intake line between the intake and the pre-filter, allowing for the sample line to reach the water.

Suspended solids are typically high at both watershed sites, so the trace organics sampler pump speed is increased to 300-400 RPM higher than the speed used for the vessel-accessed sites. Even with increased pump speed, the 20 L sample carboys typically fill in 15-20 minutes instead of the usual 10-12 minutes. The filters and/or pre-filters are cleaned and replaced as necessary to maintain a good flow of water out of the exit tube.

### 3.1.13 Water Sample Handling and Shipping

Samples for trace metals analysis, nutrients, DOC, Chlorophyll, TSS, and Chromium are maintained on board the vessel and transferred to the laboratory by UCSC personnel. The nutrient samples are frozen on dry ice on the vessel, and maintained frozen until they are transferred to laboratory freezers. All other trace metal and related samples are stored in sealed buckets at room temperature on board the vessel and transferred to the laboratory at the conclusion of the cruise. The UCSC cruise manager(s) are responsible for maintaining sample integrity throughout the cruise. Sample contamination is avoided by double bagging the sample containers, handling the containers with clean gloves, and transferring the samples into sealed buckets/coolers immediately after sampling.

Samples for organics analysis consist of particulate filters and resin filled columns. After obtaining the samples, the columns and filters are handled in the following manner: The column assembly is removed from the sampler with the following steps: the columns are removed by loosening the Gorilla grips™ with the proper wrenches. Residual water is drained from the columns, and the methanol rinsed end-caps are attached. The end-caps are wrapped with Teflon™ tape and the sample log sheets are filled out with any pertinent information. The columns are placed in a cooler with enough dry ice to keep them cool (usually 5 pounds put in every morning and every evening, depending on ambient conditions). The filter housing containing the used filter is removed with the following steps: a large pair of forceps is rinsed with methanol and used to remove the filter from the filter housing. The filter is wrapped with a piece of methanol rinsed foil, and is over-wrapped with a second piece of clean foil. The filter is labeled with the collection date, site code and the collector's initials and is then placed in a polyethylene bag which is twisted, wrapped over the filter again, and labeled with the same information on the outside of the bag. The bag is placed in a cooler containing enough dry ice to keep the samples frozen (100 pounds is usually enough to add small amounts to the column coolers and keep the filters frozen for one week of the cruise). On return to the U of U laboratory, they are placed in a freezer.

Samples for analysis of Arsenic, Selenium, and Mercury are double bagged and held in coolers chilled with water ice on board the vessel. The AMS cruise manager maintains them on the vessel, and ships them overnight to BRL in Seattle, Washington at the mid-point and end of the cruise via a commercial carrier. The As, Hg, and Se samples are packed in coolers with blue ice, and are accompanied by chain of custody form.

Water toxicity samples are collected into clean flouride coated polyethylene carboys. After obtaining the sample, each carboy is labeled with the site code, date, analysis, and the collector's initials. The samples are held on wet ice, and then picked up by PERL personnel at the dock, or delivered to their facility. Toxicity sampling information is logged on the site information sheet, and COC's are prepared for handing the samples off to PERL personnel.

The samples that are held on either wet ice or dry ice are checked periodically to ensure that samples are appropriately protected and stored. Ice is added as required. Additionally, coolers containing wet ice are drained periodically to remove water from melted ice.

In addition to the ship's log, a sample record is maintained for each site. The sample record contains the following information:

1. Station name and code
2. Cruise number and collection date
3. Arrival and departure time at each station
4. Station coordinates (latitude and longitude)
5. Water depth at time of sampling
6. A record of every sample bottle filled, with bottle identification code number and quantity of bottles
7. Collecting personnel's names
8. Water temperature
9. Salinity
10. CTD file name
11. CTD start time
12. Other remarks (i.e. any conditions that could possibly influence sample analysis or data interpretation)

The sample collection form, coupled with a chain of custody record and a laboratory analysis record, allows tracing of the complete history of a sample from time of collection to final entry of data to a computer database.

At the conclusion of each cruise, UU personnel maintain possession of trace organics samples and UCSC personnel maintain possession of trace elements samples. All other samples are shipped by AMS personnel with COC's to the appropriate laboratories for analysis. Water samples are shipped overnight in coolers on enough blue ice to maintain a cool environment for two days. The receiving laboratories are requested to notify AMS of receipt of samples and forward completed COC's to SFEI.

## 3.2 Sediment Sampling

### 3.2.1 Overview and Objectives

Sediment sampling for the RMP consists of 26 stations within the Estuary, including two watershed stations. Sampling normally takes six working days. Four laboratories perform sediment analyses of trace elements, total organic carbon, grain size and organic compounds. In addition, sediment toxicity bioassays are conducted at 14 stations and pore water chemistry (pH and ammonia) is processed aboard the vessel at all sites. Two grabs are collected from each station and composited to produce a single sample that is partitioned according to the various analyses.

Collecting sediment samples is problematic. Samples of surficial sediments (top 5 cm) for analysis of chemical constituents must be collected in a manner such that surface layers are not disrupted when removed from the bottom of the sample for processing. Disruption may cause mixing of surficial layers with lower layers in the sample, and may lead to dilution or concentration of the contaminants of concern, depending upon the chemical content of the various layers of sediment.

Sediment sampling is performed using a Young-modified, Van Veen grab with a surface area of 0.1 m<sup>2</sup>. The grab is constructed entirely of stainless steel and the jaws and doors are coated with Kynar™ to improve chemical inertness. A scoop and bucket used to remove and composite sediments are also constructed of stainless steel and coated with Kynar™.

A minimum of five cruise participants (excluding vessel captain) are required to conduct a sediment cruise, although six participants are recommended. Specific responsibilities and assignments for cruise participants is presented in Table 12.

**Table 12. Crew responsibilities for RMP sediment cruise.**

<i>Cruise Participants (Subcontractors)</i>	<i>Responsibilities</i>
Applied Marine Sciences, Inc. (AMS)	Cruise management, sediment chemistry sample collection, sediment and pore water chemistry, CTD operation and watershed sampling
City and County of San Francisco (CCSF)	
University of California, Santa Cruz (UCSC)	Vessel operation
San Francisco Estuary Institute (SFEI)	Sample collection

AMS is responsible for oversight of sampling operations, compliance with cruise plan and quality assurance guidelines, maintenance of the sample field log and chain-of-custody records, and operation of the CTD. CCSF is responsible for benthic sampling and processing of benthic samples. UCSC is responsible for vessel operation and safety. SFEI will be responsible for providing one or two technicians to assist in sample collections and processing.

It is the responsibility of AMS to ensure that all field personnel are capable of sampling safely and complying with the quality assurance guidelines. AMS is required to ensure that:

- Field personnel understand and follow the vessel operating safety procedures as described by the vessel captain. Any concern or uncertainty about operational procedures or safety practices must be brought to the attention of the vessel captain or the AMS cruise manager immediately.
- Field personnel will strictly adhere to the quality assurance protocols (see QAPP plan) to insure the collection of representative, uncontaminated sediment chemistry samples.
- Field personnel are thoroughly trained in the proper use of sample collection gear and are able to distinguish acceptable versus unacceptable samples in accordance with pre-established criteria.
- Field personnel are thoroughly trained to recognize and avoid potential sources of sample contamination (e.g., engine exhaust, winch wires, deck surfaces, ice used for cooling samples, etc.).
- Field personnel follow established procedures for sample collection, processing, documentation, and distribution.
- Field personnel make use of appropriate personal safety equipment at the discretion of the cruise manager or vessel skipper.

The objectives of the sediment cruise are:

Collect sediment samples at 26 stations for the analysis of:

- Trace metals and trace organics by the Bay Area Dischargers Authority (BADA)
- As, Hg, Se by Brooks-Rand (BRL)
- Grain size, TOC and total nitrogen by the University of California, Santa Cruz (UCSC)
- Pore water pH and ammonia by Applied Marine Sciences (AMS)
- Pore water sulfides by Marine Pollution Studies Lab (MPSL)
- Foraminifera by US Geological Survey (USGS)
- CTD profiles by Applied Marine Sciences (AMS)
- Collect sediment samples at 14 stations for analysis of toxicity by MPSL
- Collect sediment samples at nine stations for analysis of benthic infauna by City and County of San Francisco (CCSF).

It is critical that sample contamination be avoided during collection. All sampling equipment (i.e., Van Veen grab, compositing bucket and scoops) are composed of a non-contaminating material and are thoroughly cleaned before each use. Sampling personnel wear polyethylene gloves whenever taking or processing samples to avoid contact contamination. In addition, airborne contamination is avoided by keeping sample containers, sample scoops and compositing bucket inside the vessel cabin with door closed or appropriately covered when not in use.

### **3.2.2 Sediment Sampling Vessel Safety**

There are 26 sediment sampling sites currently in use by the RMP. Twenty four sites are sampled from a survey vessel and two sites (watershed sites) are sampled from land. All sites

except for the watershed sites are sampled from the *R/V David Johnston*. Important features that make the *David Johnston* well-suited for sediment cruises include a large deck, large enclosed cabin with work benches, and an A-frame for deploying and retrieving the Van Veen and Ponar grabs. The watershed sites (Guadalupe River and Standish Dam) are sampled by car due to inaccessibility by boat.

The maximum work day cannot exceed 12 hours per United States Coast Guard requirements. The captain reserves the right to cancel or modify the cruise for any safety reason that could endanger customer, crew or captain's safety. Captain reserves the right to modify any procedure that could damage the vessel. Customer and any personnel brought aboard by customer agree to follow all safety procedures and policies implemented by the captain. Any areas of concern for crew members should be brought to the immediate attention of either the cruise manager or the vessel captain.

General safety procedures used on the sediment cruise involve the following guidelines:

1. Boating Safety - A safety briefing is given by the skipper of the vessel prior to departure from the dock. This briefing shall include at a minimum the location of flotation devices, location of fire extinguishers, and emergency procedures. Enough information is given to allow the crew to communicate with emergency services, and operate the vessel in the event that the captain is incapacitated.
2. Equipment Storage - All equipment to be used is properly stowed to minimize the possibility of movement during vessel transit.
3. Arrival at Sampling Site - Sampling cruise personnel assist the skipper in setting the anchor at each sampling site.
4. Winch Operation - The vessel captain operates the winch that deploys and retrieves the grabs. The vessel captain may decline to use the grabs when rough conditions preclude their use.
5. Chemicals - Buffered formalin and dilute solutions of HCl and methanol are used as part of the sampling operations. Personal protective gear is provided for use by field crew, including eye protection, foul-weather gear, and gloves.

### **3.2.3 Sediment Sampling Equipment List**

Preparation of sediment sampling equipment is the responsibility of AMS and begins at least four days before each cruise. The CTD and pore water sampling equipment requires special advance preparation, calibration and cleaning. An equipment list for sediment sampling (not including pore water analysis equipment and sample containers) is provided in Table 13.

**Table 13. Equipment list for sediment sampling.**

<i>Quantity</i>	<i>Description</i>
1	Van Veen grab, 0.1 square meter capacity, Kynar™ coated, (pre-cleaned in the laboratory)
1	Van Veen grab stand
2	Plastic floats for Van Veen grab
2	Weights for Van Veen grab
1	SBE1 19 CTD, calibrated in the laboratory prior to use
1	Data terminal and communication cable for CTD
1	CTD Maintenance kit including 8 new “D” size batteries
1	Sediment overflow bucket
8	Insulated plastic coolers for sample storage, (pre-cleaned)
1	Keys to Coyote Creek gate (for watershed sampling)
60-90 (lbs.)	dry ice
1	Insulated plastic cooler for dry ice storage
1 (pr.)	Cotton gloves for dry ice handling
1	Cruise plan
30	Sample collection forms
10	Chain of custody forms
30	Pore water collection forms
2	Label tape
2	Aluminum foil, 100 square feet
48	Ziploc™ bags, 1 gallon size
2	Sharpie pens, thin and wide
200	Latex gloves, non-powdered
1	Splash-proof eye protection
2	Plastic brushes
3	Five gallon plastic buckets
3	Hydrochloric acid 10%, 4 L amber bottle, reagent grade
3	Methanol, 4 L amber bottle, reagent grade
5	Deionized/reverse osmosis water, 4 L polyethylene bottle
1	Alconox™ detergent
2	Plastic floats
3	Teflon™ squeeze bottles, (pre-cleaned) in the laboratory (labeled for distilled water, 1%hydrochloric acid and methanol)
3	Kynar™ coated scoops, (pre-cleaned) in the laboratory
1	Kynar™ coated bucket, (pre-cleaned) in the laboratory
1	Cellular phone with battery charger
as needed	Formalin gloves
as needed	Preprinted sample labels
as needed	Paper and cloth towels
as needed	Foul weather gear for splash protection, boots

### 3.2.4 Sample Containers

The sample containers used for sediment samples and the laboratory responsible for providing them are listed in Table 14. Each container is given a permanent sample label written in waterproof ink. At a minimum, each sample label includes station name and code, sample date, collection time, analysis required, and collector's initials.

Sample containers are cleaned and prepared by the analyzing laboratory, or are factory pre-cleaned, and are delivered to AMS at least one week prior to the start of a cruise. Sample containers are pre-labeled and packed into pre-cleaned Igloo™ coolers. A container list is prepared before a cruise starts and is used to verify that all samples are properly collected in the field. At least two personnel verify that the proper sample containers for each station have been filled with sediment and that the labels correspond to the proper station name and code.

**Table 14. Container list for sediment sampling.**

<i>Sample Type</i>	<i>Container</i>
As, Hg, Se	18 ml Teflon™ jar, pre-cleaned by BRL. Fill with sediments to 0.25" from top.
Trace Organics	New 100 ml Ichem™, wide-mouth, glass with Teflon™ liner, certified trace organics clean by I-Chem™ and provided by BADA. Fill with sediments to 1" from top. Do not overfill.
Trace Elements	New 60 ml Nalgene™ polyethylene jar, certified trace metal clean by Nalgene™ and provided by BADA. Fill with sediments to top.
Archive	Same as trace organics container. Fill with sediments to 1" from top.
Pore Water Sulfides	4.5 ml glass scintillation vial, pre-cleaned by MPSL and provided by MPSL. Fill with pore water to top, leave no head-space.
Toxicity	1 L glass I-chem™ wide-mouth jars, certified trace organics clean by I-Chem™ and provided by MPSL. Fill with sediments to top, leave no head-space.
Benthic Infauna	500 ml glass jar, pre-cleaned and provided by CCSF.
pH and Ammonia	On-board measurements only
Foraminifera	250 ml glass jars prepared with formaldehyde and rose bengal solution by USGS

### 3.2.5 Sediment Sampling Equipment Preparation

Sediment sampling equipment is prepared in the laboratory by AMS a minimum of four days prior to the start of a cruise. The sampling equipment that is pre-cleaned include:

- Van Veen Grab (excluding frame and stand)
- Sample scoops (three)
- Compositing bucket
- Wash bottles
- Glass pore water coring tubes (six)

Use the following procedures for cleaning sediment sampling equipment:

1. Soak equipment (fully immersed) for three days in a 0.5 % solution of Alconox™ detergent and deionized water.
2. Rinse equipment three times with deionized water and let dry in a clean place.
3. Rinse equipment with 1.0 % solution of hydrochloric acid, followed by a rinse with petroleum ether, followed by another set of three rinses with deionized water. All equipment is then allowed dry in a clean place.

The cleaned grab is wrapped in aluminum foil until used in the field. All other equipment is stored in clean Ziploc™ bags until used in the field.

The CTD is checked for proper operation at least 48 hours before use. Refer to the section on CTD maintenance and calibration in the Water Sampling section (3.1.5.11). The pore

water ammonia probe must be assembled and calibrated at least 24 hours before use. Refer to the section on pore water sampling (3.2.6.3) for assembly instructions.

### 3.2.6 Sediment Sampling Procedures

When the vessel reaches a sampling station and the anchor has been deployed, the captain notifies personnel that the vessel is on site and switches on a bilge pump used for rinsing the sampling equipment. Sampling equipment is cleaned at each station using the following methods:

1. Fill the compositing bucket with Estuary water from the raw water pump and add approximately 1/8 cup of Alconox™ detergent to the bucket.
2. Place all sampling scoops and glass coring tubes into the bucket and wash thoroughly with the Alconox™ solution. Wash all Kynar™-coated parts of the Van Veen grab with Alconox™ solution.
3. Completely rinse the grab, bucket, sample scoops and coring tubes with Estuary water.
4. Rinse the grab, bucket, sample scoops and coring tubes with 1.0 % HCl followed with a rinse of methanol.
5. Completely rinse the grab, bucket, sample scoops and coring tubes with deionized water and let air dry. Cover all cleaned parts with aluminum foil until use.

At stations where benthic sampling is conducted, a Ponar grab is used to collect one acceptable benthic sample prior to using the Van Veen grab for collecting sediment chemistry samples. Personnel from CCSF are responsible for operating the Ponar Grab and determining if grabs taken meet acceptance criteria. After benthic samples have been collected, the Ponar grab is replaced on the hydrowire with the Van Veen grab for collection of chemistry samples. Two grabs are taken at each site. If the sediments at a station are considerably fine, plastic floats may be attached to the grab frame and secured so they do not interfere with grab operation. Likewise, if the sediments are considerably coarse, weights are added to the grab frame to assist penetration of the sediments. The quality of grab samples is ensured by requiring each sample to satisfy acceptance criteria concerning the depth of penetration and disturbance of the sediment within the grab.

Samples contain only the top 5-cm of sediment within the area of the grab jaws. Samples are rejected under the following conditions:

1. There is a rock or shell fragment wedged between the jaws of the grab allowing the sample to wash out.
2. The sample surface is significantly disturbed.
3. The sample is uneven from side to side, indicating that the grab was tilted when it penetrated the sediment.
4. The surface of the sample is in contact with the doors of the grab, indicating over-penetration of the grab and possible loss of material around the doors.

After determining a grab meets acceptance criteria, overlying water is drained off. Using pre-cleaned glass cores, three 5-cm deep cores are taken from each side of the grab. These cores are used for measurement of pH, ammonia, and total sulfides in pore water.

If sampling of foraminifera is required, 20-30 grams of surficial sediment is removed from one grab. Since foraminifera analysis does not require compositing, sediment can be used from grabs that otherwise would not pass acceptance criteria for sediment chemistry analysis. USGS supplies coolers and sample containers containing a formalin solution and rose bengal for preservation of foram samples. Sediment for foram analysis is taken from the top one inch of the grab only and placed in an intermediate container with no preservative. The intermediate container is capped and set aside until all other allocation of sediment for chemical analysis is completed. This is done to minimize the potential for contamination of samples by the formalin used to preserve the foraminifera. After all other required procedures are completed, the sediment for foraminifera analysis transferred to the proper containers.

The remaining top 5-cm of sediment is scooped from each of two replicate grabs and mixed in the compositing bucket to provide a single composite sample from each site. Portions of the composited sample are placed into clean containers provided by each laboratory. A duplicate chemistry sample is collected from the composite for archiving and is labeled as an “archive”. Cores collected for analysis of pore water are centrifuged onboard the vessel. Part of the supernatant is then used for analysis of ammonia and pH (performed on-board the vessel by AMS) and part is preserved for analysis of sulfides (analyzed in the laboratory by MPSL)

### **3.2.6.1 Sediment Chemistry**

There is no on-board sediment chemistry except for analysis of pore water pH and ammonia. Fill sample chemistry containers according to instructions given in the container list (Table 14). Refer to section 3.2.6.3 for analysis of pore water pH and ammonia.

### **3.2.6.2 Benthic Infauna**

Benthic infauna primarily comprises sedentary, invertebrate organisms that burrow in or live on the surface of sediments. Benthic infauna communities fluctuate in response to natural and human induced environmental perturbations and therefore can be important indicators of environmental health. For this reason they often are an important component of many ecological monitoring programs. Benthic infauna is sampled with a Ponar grab with a surface area of 0.05 square meters. The grab is equipped with hinged stainless steel mesh lids with rubber flaps to allow flow-through of water during descent and thus minimize disturbance of surface sediments. The rubber flaps close upon retrieval and prevent winnowing of the sample.

One infauna sample is collected at nine RMP sediment stations. Sampling procedures will insure that samples are collected from a localized area at each station to reduce uncontrolled temporal and spatial variations. Lead weights are added to or removed from the outside of the grab as appropriate for sediment type to control depth of penetration.

After deployment and retrieval, the grab is placed on a stand for processing. The grab lids are opened and the sample is examined for suitability using the following criteria:

- Complete closure of the grab jaws.
- No evidence of sediment washout through the grab doors.

- An even distribution of the sediment in the grab.
- Minimum disturbance of the sediment surface.
- Minimum overall sediment depth appropriate for the sediment type: 4 cm in coarse sands and gravel, 5 cm in medium sands, 7 cm in fine sands, and 10 cm in silty sands, silts, and clay.

If the sample passes all of the criteria, the grab jaws are opened and the sample is dumped into a five gallon plastic bucket placed beneath the grab stand. Estuary water is used to wash all sediment from the grab and grab stand into the bucket. Care is exercised not to lose sediment by overfilling the bucket. The sample bucket is then moved to a wash table for sample sieving.

When a sample bucket arrives at the sieving station, it is lifted to the sieve table and poured slowly onto the nested sieve screens. The sea water hose with a flow control nozzle is used to slowly wash sediment from the sample bucket onto the sieve screens. The sieving process is aided by keeping sediment in suspension as it reaches the screen. The sample is washed from the sample bucket until the bucket is empty and well rinsed. Sediment is washed through the nested sieve screens by gently running seawater over the top screen. Use of high water pressure damages organisms impinged on the sieve screen mesh.

When all material smaller than 1.0 mm has passed through the top screen, the process is repeated with the finer screen until all material smaller than 0.5 mm has passed through. The material retained on each screen is gently washed into one corner of the screen and with the aid of a canning funnel, washed into separate appropriately labeled sample jars. A wash bottle with seawater is used to rinse any material on the inside screen frame and canning funnel into the sample jar. Any organisms remaining on the screens are carefully removed with forceps and placed in the appropriate sample jars. The sample jars are then capped with dome lids and bands, labeled with indelible ink inside and out, and delivered to the on-board formalin station. Great care is exercised to avoid creating fragments when removing organisms from the sieve screens. The sieve screens are rinsed with high-pressure seawater and scrubbed clean with a stiff-bristle brush between samples.

If the sample contains many shell fragments and/or worm tubes, the sediment sample is added to the top (1.0 mm) screen in stages so that the screen does not become too full. If the bottom screen (0.5 mm) begins to clog with sediment, the field crew ceases adding sample and gently runs the hose nozzle with low flow along the outside bottom of the 0.5 mm screen being careful not to lose sample by allowing water to escape over the top of the sieve. The material retained on a sieve screen is not allowed to fill the sample jar more than half full. In such a case, the material is divided among two or more jars and each jar is labeled as jar 1 of 2, jar 2 of 2, etc., as required.

At the formalin station, each sample jar lid is replaced with screen lids fitted with 0.25 mm Nitex™ mesh and the Estuary water is decanted from the sample jars through the screen lids. Relaxant (isotonic MgCl<sub>2</sub>) is added to the sample through the screen lid to a level approximately one third higher than the sample level. A wash bottle of relaxant is used to wash down the screen lid and sides of the sample jar. The sample jar is recapped with the sample jar lid and gently rotated several times in a tilted position to ensure mixing of the relaxant throughout the sample. The sample is allowed to sit in the relaxant for 15-30 minutes. After this

period, the sample jar lid is replaced with a screen lid and the  $MgCl_2$  is decanted out of the sample jar in preparation for fixing the sample.

At the formalin station, relaxant is decanted out and fixative (10% buffered formalin in seawater) is added to the sample through the screen lid. Fixative is added to a level approximately one third higher than the sample level. A wash bottle of fixative is used to wash down the screen lid and sides of the sample jar. The screen lid is removed, 2 or 3 drops of stain (rose bengal solution) are added to the sample and the sample jar is recapped with the sample jar lid. The jar is gently rotated several times in a tilted position to ensure mixing of the fixative and stain with the sample. Safety glasses and nitrile gloves are worn when working with fixative.

While onboard the survey vessel, benthic infauna samples are stored in plastic trays with dividers, then transferred to cardboard cartons with dividers for travel to the laboratory for sample sorting. Benthic infauna samples fixed in formalin are washed in tap water and transferred to 70% ethyl alcohol between 24 and 72 hours after fixation. Samples can then be held indefinitely in 70% ethyl alcohol.

A sample collection log, maintained by CCSF, records sample date, station, depth of grab penetration, number of grabs, number of bottles per sample, and any problems encountered.

### **3.2.6.3 Sediment Pore Water Analysis**

Pore water samples removed from benthic grabs are analyzed for pH, ammonia and sulfides. Ammonia and sulfides are a natural component of marine sediments and common constituent of municipal effluents. Ammonia and sulfides may occur in concentrations that are toxic to marine organisms in toxicity tests and, therefore, must be accurately measured.

Table 15 presents the equipment list for pore water analysis. This equipment is prepared in the laboratory at least four days prior to the start of each cruise.

**Table 15. Equipment list for pore water sampling.**

<i>Quantity</i>	<i>Description</i>
1	Orion™ model 95-12 selective ion ammonia electrode
1	Corning™ model 240 general purpose, gel-filled pH probe
1	Corning™ Model 240 temperature compensated pH/millivolt meter with temperature probe
1	IEC™ “Clinical” centrifuge with fixed angle rotor, 6 x 50 ml capacity, fitted with stainless steel sleeves
200	50 ml centrifuge tubes with screw caps
1	Magnetic stir plate
3	Micro-magnetic stir bars
1	Electrolyte solution for pH and ammonia electrodes
1	60 ml internal filling solution for ammonia probe
1	Membrane kit for ammonia probe
1	250 ml standardizing solution, NH <sub>4</sub> -Cl, 1,000 ppm as N
3	Standards for pH, 4.0, 7.0 and 10.0, NIST traceable
4	Standard reference solutions of 0.1, 1.0, 5.0 and 10.0 ppm ammonia prepared as necessary from a stock solution of 1,000 ppm ammonia certified reference material
1	Ionic strength adjuster (ISA)
4	4-cycle semi-log paper for graphing calibration curve
1	Compressed nitrogen tank with regulator and hose
4	Glass coring tubes with rubber stoppers, 3.5 cm diameter,
1	Centrifuge tube rack
1	100 ml flask, for making ammonia standards
3	10 ml beakers
1	Eppendorf™ pipette, 100 ul capacity with spare tips
2	25 ml glass serological pipettes
30	4.5 ml glass scintillation vials, screw-cap with plastic liner, prepared with zinc acetate preservative (see pore water sulfides procedures)
4	Distilled water, 4 liters, processed by reverse osmosis and de-ionization, ammonia free
1	Record book
1	10 ml pipette
as needed	Kimwipes™

The following procedures outline the steps necessary for preparation of the pore water electrode. Electrode calibration is performed prior to the start of the sediment sampling cruise and whenever the electrode operation becomes suspect

### **Pore Water Electrode Preparation**

This procedure is used to set up the electrode prior to calibration. For best results, the electrode should be prepared at least 24 hours before calibration:

1. Unscrew the cap and remove the glass inner body from the outer body. Set the cap with inner body aside carefully.
2. Remove the bottom cap from the electrode outer body.
3. Using tweezers, remove the old ammonia membrane then carefully grasp the edge of a new membrane from between paper separators.

4. Place the membrane against the threads of the probe outer body so that one of the shorter sides of the membrane is in line with the threads. Loosely stretch the membrane lengthwise across the opening holding the edge with your fingers.
5. Replace the membrane cap and screw on until finger-tight. The membrane must be smooth with no wrinkles.
6. Fill the outer body with electrode filling solution.
7. Place the inner body into the outer body containing internal filling solution and screw on the upper cap.
8. Shake the fully assembled electrode as if it were a clinical thermometer to remove bubbles.

### **Pore Water Electrode Calibration**

This procedure measures electrode slope, used for calibration of the pore water electrode. Slope is defined as the change in millivolt potential observed with every tenfold change in ammonia concentration:

1. If the electrode has been stored dry, prepare the electrode as described above.
2. Connect the electrode to the meter.
3. Place 100 ml of distilled water into a 150 ml beaker. Add 2 ml ionic strength adjuster (ISA). Stir thoroughly. Set the function switch of the meter to the mV mode.
4. Rinse the electrodes with distilled water and place in the solution prepared in the previous step. To prevent air entrapment on the membrane surface, be sure to use an electrode holder that keeps the electrode at a 20° angle. If bubbles appear on the sensing membrane, tap electrode gently to remove them.
5. Pipette 1 ml of the 1,000 ppm ammonium chloride standard into the beaker and stir thoroughly. When a stable reading is displayed, record the electrode potential in millivolt.
6. Pipette 10 ml of the same standard into the same beaker. When a stable reading is displayed, record the electrode potential in millivolts.
7. The difference between the first and second potential reading is defined as the slope of the electrode. The difference should be in the range of -54 to -60 mV/decade when the solution temperature is between 20-25 °C. Record the temperature of the ammonia standard for future reference.

Ammonia electrode calibration is accomplished by direct measurement with at least three standards (10.0, 1.0, 0.1 ppm). Blank correction is not necessary. Use the following steps for electrode calibration:

1. Set the function switch of the pH/millivolt meter to mV mode.
2. Make the standards by placing 10 pL, 100 pL, and 1,000 pL of the 1,000 ppm nitrogen solution into each of three Erlenmeyer™ flasks.
3. To each flask add enough distilled water to make 100 ml of solution.
4. Add 2.0 ml ISA to each flask and stir thoroughly..
5. Measure the millivolt potential of the 0.1 standard. Stir thoroughly until the meter displays a constant measurement. Record the reading. Repeat process with the 1.0 ppm and 10.0 ppm standards, rinsing with DI water and blotting dry between standards. Using log 10 graph paper, generate a calibration curve from the millivolt readings obtained.

## Pore Water Sample Collection

Use the following guidelines for pore water sample collection:

1. Wash coring tubes using the same procedures as outlined for preparation of the Van Veen grab and wrap in clean aluminum foil.
2. After retrieving the Van Veen grab and determining the sample suitability, insert a coring tube into the sediment so that there is 5.0 cm of sediment in the tube after it has been withdrawn. Three cores are removed from each side of the grab for a total of six cores taken per sampling site. Coring tubes may be reused at the same station without washing.
3. Place the sediment from each core into a separate clean centrifuge tube and transport the tubes into the vessel cabin. Purge the headspace of each tube with nitrogen then cap each tube tightly.
4. Place the six centrifuge tubes into the centrifuge and spin at a rate of 400-1,000 RPM. Be aware that the centrifuge may transfer excessive heat to the sample. If this occurs, the sample must be chilled to the temperature of the calibration solution of the ammonia probe.
5. Rinse a collection pipette with DI water and flush it dry.
6. Pipette out all the pore water from the top of each of the six centrifuge tubes into one empty centrifuge tube.
7. Pipette out enough pore water from the composited centrifuge tube to fill the sulfides vial with no headspace or bubbles. This requires about 1.5-2.0 ml of pore water. Do not spill the preservative in the sulfide bottle and do not contact the pipette with the sample water in the sulfides vial (it contains acid and could corrupt ammonia measurements). Record the station code and date on the sulfides vial and store it in a dark place (do not freeze the sample).
8. Pipette 10.0 ml of supernatant into a small beaker, place a clean micro stir rod into the beaker and gently stir. Do not add ISA.
9. Insert the pH probe and tip of the thermocouple probe into the sample. Record the pH of the sample without adding ISA.
10. Add approximately 0.02 ml of ISA per ml of sample (about 0.2 ml ISA in a 10.0 ml sample) or enough ISA to bring the pH to 11 or above.
11. Disconnect the pH probe from the meter and install the ammonia probe. Rinse the ammonia probe with deionized water and blot dry with a clean towel before placing it into the sample. Record the millivolt reading when the meter stabilizes. This normally takes about four minutes for a sample of low ammonia concentration (less than 1.0 ppm).
12. Plot the mV reading from the meter of the unknown sample concentration with the curve generated from the known standards to find the ammonia concentration of the sample.

For overnight or weekly storage, the electrode tip should be immersed in a 1,000 ppm standard  $\text{NH}_3$  solution without added ISA. For storage over one week, completely disassemble the electrode and rinse the inner body, outer body, and bottom cap with distilled water. Dry and reassemble electrode without filling solution or membrane.

If the electrode is accidentally left in the air and erratic results are obtained, the space between the inside of the membrane and the sensing element may be dry. To make the electrode usable again, withdraw the glass electrode from the membrane by pulling back the cable slightly. New solution will flow into the space.

#### **3.2.6.4 Sediment CTD Profiling**

A CTD cast is recorded at each station, except for the two watershed sites. The procedures for operating the CTD on the sediment cruise are the same as those for the water cruise. Refer to section 3.1.10 (Water CTD Profiling) for CTD operating instructions.

#### **3.2.6.5 Watershed Sediment Sampling**

All equipment is pre-cleaned according to the procedures as outlined above. The guidelines for obtaining grab samples at the watershed sites are as follows:

1. Randomly select an area of unconsolidated fine-grain sediment. Unconsolidated sediments lack a usually visible diatom covering and are very easily penetrated. Typical locations are the side slope or surface of recent slump blocks and the surface of actively accreting point bars on the inside of meander bends. For drainage divide stations, randomly select a location at least 10 meters from any channel or ditch, and at least 5 meters from the upland edge of the tidal marsh. Do not select spots in ponds or channel pans.
2. Insert a cleaned scoop into the sediments to a depth of 5 centimeters. Remove sediments from an area approximately 0.1 square meter. The total amount of sediment sampled is proportional to the amount of sediments removed when using the Van Veen grab at the Estuary sites.
3. Place sediment into a pre-cleaned compositing bucket. Thoroughly stir the combined material into one homogeneous mixture.
4. Place the appropriate amounts of the sediment into clean containers with appropriate labels, and place the containers on ice for short-term storage.
5. To avoid cross-contamination between stations, all utensils, buckets, and the glass core tubes must be rinsed between stations with Estuary water, then scrubbed thoroughly with Alconox™, followed successively by one rinse with deionized water, one rinse with 1% HCl, one rinse with methanol, and a final rinse with deionized water.

#### **3.2.6.6 Sediment Sample Storage and Handling**

Samples are stored aboard the vessel according to the type of analysis performed. Normally, samples used for chemistry analysis are stored on dry ice while samples used for toxicity analysis are stored on wet ice. On board the vessel, properly labeled sample containers are segregated by analysis required and stored according to the guidelines listed in Table 16.

**Table 16. Storage methods for sediment samples.**

<i>Analysis</i>	<i>Method of Storage</i>
Trace Elements	Store on dry ice in Igloo™ cooler
Organics	Store on dry ice in Igloo™ cooler
Arsenic, Selenium, Mercury	Store on dry ice in Igloo™ cooler, keep in separate Ziploc™ bags
Cognates	Store on dry ice in Igloo™ cooler
Archives	Store on dry ice in Igloo™ cooler
Toxicity	Store on wet ice in Igloo™ cooler, replenish ice each day
Benthic Infauna	Preserved, no chilling required
Foraminifera	Preserved, no chilling required
Sulfides	Stored in a dark location

The samples on both wet ice and dry ice should be checked periodically to ensure that samples are appropriately protected and ice should be added as required. Additionally, coolers containing wet ice should be drained periodically to remove melt water.

In addition to the ship's log, a sample record is maintained for each site. The sample record contains the following information:

1. Station name and code
2. Collection date
3. Arrival and departure time at each station
4. Station coordinates (latitude and longitude) from the survey vessel's GPS
5. Depth at time of sampling from the ship's depth meter
6. A record of every sample bottle filled, with bottle identification code and quantity
7. Collecting personnel
8. Other remarks (i.e. any conditions that could possibly influence sample analysis or data interpretation or notation of the general performance of equipment involved with the sampling.)

The sample collection form, coupled with a chain of custody record and a laboratory analysis record, allows tracing of the complete history of a sample from time of collection to final entry of data to a computer database. In addition to the sample collection form, some of the laboratories may use a bottle labeling system that catalogs the preparation of bottles prior to their use in the field. This system is particularly important for the Teflon™ bottles used in trace element analysis, where exhaustive cleaning procedures are employed before releasing them for field sampling.

### **3.3 Bioaccumulation Sampling**

#### **3.3.1 Overview and Objectives**

The bioaccumulation study of the RMP is conducted to document long-term trends in bioavailable contaminants in the Estuary. Bioaccumulation sites are chosen to incorporate contaminant sources from broad regions of the Estuary and reduce effects from specific point sources.

Bioaccumulation sampling consists of collecting oysters (*Crassostrea gigas*) and mussels (*Mytilus californianus*) from “background sites” of known chemistry and deploying the bivalves at 13 locations in the Estuary. Resident clams (*Corbicula fluminea*) are also collected from two sites on the Sacramento and San Joaquin rivers. Bivalves are deployed twice each year, usually in January and June. The two deployment periods represent the “wet season” and “dry season”, respectively. Deployment duration is approximately 100 days with a “maintenance” cruise occurring approximately 50 days after deployment.

Analysis of contaminant concentrations is conducted on a subset of the transplanted bivalves prior to deployment (time zero) in Estuary locations, and after deployment. The differences between pre- and post-deployment contaminant concentrations allow determination of contaminant uptake during the period of deployment. Transplanted bivalves (time one) are also collected from the time zero collection sites at the end of the deployment period, to obtain information on uptake variables affecting wild populations during the deployment period.

Bivalve condition is also measured as part of the bioaccumulation study. Condition is the ratio of dry tissue weight to shell cavity volume. Bivalves have high condition when there is abundant food and few environmental or physiologic stressors.

#### **3.3.2 Oyster Collection**

*Crassostrea gigas* are purchased from a commercial grower located in Tomales Bay, CA. A total of 1200 “extra small” (grower’s designation) oysters are obtained from the grower by AMS personnel and transported in coolers to the Bodega Marine Laboratory (BML) in Bodega Bay, CA.

The oysters are placed into 36" polypropylene mesh bags containing 36 oysters divided into three sections of twelve each; the three sections are separated by nylon cable ties woven through and around the outside of each bag. Room is left in each section to allow for growth and movement of the oysters within the bag.

The bags of oysters are placed on racks in an outdoor tank supplied with aeration and freely flowing, full strength seawater. The oysters are maintained in this tank for a maximum of two weeks prior to deployment. They are transported from BML to the vessel in coolers which have been scrubbed with Alconox™ detergent, rinsed with fresh water and chilled with freshwater ice. During transportation, the coolers are allowed to drain, preventing the oysters from being submerged in fresh water from the melting ice.

### **3.3.3 Mussel Collection**

*Mytilus californianus* are collected from a location that is several hundred yards north of the parking lot at Bodega Head, near Bodega Bay, CA. Collection of mussels is done by 3-4 AMS personnel, all of whom have in possession valid California Department of Fish and Game (CDF&G) scientific collecting permits. The CDF&G is notified of mussel collection efforts 24 hours in advance. Additionally, AMS has a permit from the Sonoma County Division of State Parks, who also require a scientific collecting permit and notification prior to collection. When weather conditions are marginal for safe sampling and as deemed necessary by AMS personnel, individuals employed BML serve as wave spotters, to warn mussel collectors of potentially dangerous incoming waves.

Timing of collection efforts is determined by occurrence of a suitable low tide during daylight hours within two weeks of the beginning of the deployment cruise. Tide levels below 0 feet mean low water (minus tides) are preferred, however mussel collection may be accomplished on higher tides in calm conditions.

Safety gear used is determined by AMS personnel prior to collecting. Safety gear includes: a PFD for each individual, a life ring for emergency rescue, whistles, a hand-held radio or cellular phone, and personal foul weather gear. All personnel monitor incoming waves while collecting, and call out warnings to the other collectors if danger is perceived.

Collectors wear powder free latex gloves and collect mussels which are between 55 and 65 mm in total length. The mussels are counted as they are collected and placed into clean buckets or coolers. Approximately 1,400 mussels are collected during each of the two yearly collection efforts.

Mussels are transported to BML and placed into 40" mesh bags. Each bag contains 40 mussels each, divided into four sections of 10 mussels and separated by nylon cable ties woven through and around the bag. Room is left in each bag section to allow for growth and movement of the mussels within the bags. The bags of mussels are placed in the same tank as the oysters, and are thereafter handled and transported in the same manner.

### **3.3.4 Resident Clam Collection**

Due to a lack of viable *Corbicula fluminea* populations noted in 1998, resident bivalves are now used as a standard element of the bioaccumulation study. Clams are collected from near the historical transplantation sites in the Sacramento and San Joaquin rivers. Oysters are also transplanted at the Grizzly Bay site due to the lack of transplantable clams.

Resident clams are collected by clam dredge. The dredge is approximately two feet wide by three feet long and weighs approximately 50 pounds. The dredge is deployed from a boat using a powered winch and is dragged along the bottom. When brought to the surface, the clams are placed into a clean plastic container and packaged for analysis.

### **3.3.5 Vessel Safety**

There are 15 bioaccumulation sites in use by the RMP (refer to Table 6 for a complete list of bioaccumulation sites). Thirteen sites are bivalve transplantation sites and two are resident bivalve collection sites. All sites except for Davis Point are sampled from the *R/V Questuary*. The *R/V Questuary* is 36 feet in length and has a cruising speed of 15 knots. Important features that make it suitable for a bioaccumulation cruise include a large deck, enclosed cabin, removable swim step and an A-frame for deploying and retrieving clam sampling gear. The Davis Point site is sampled from the vessel *M.E. II*. The *M.E. II* is used because low overhead clearance is problematic at Davis Point and requires a smaller vessel for safe access.

Cruise plans are developed between AMS and the vessel skipper to ensure boat availability and that excessive currents would not pose hazards to divers. Cruise schedules may vary due to inclement weather.

It is the responsibility of the vessel captain to navigate the vessel to the bioaccumulation sites. All bioaccumulation sites are associated with fixed markers (channel markers or pilings) for easy navigation. Once at a bioaccumulation site, the vessel captain instructs the crew to fasten the vessel to the marker. At some sites, “live-boating” is conducted and the vessel is not fastened to the marker. Securing the vessel to fixed structures can pose a safety concern, thus it is the responsibility of the vessel captain to assess the safety of each site before mooring the vessel.

The maximum work day cannot exceed 12 hours per United States Coast Guard requirements. The maximum number of passengers allowed onboard the *R/V Questuary* is six, excluding captain and crew. The captain reserves the right to cancel or modify the cruise for any safety reason that could endanger customer, crew or captain’s safety. Captain reserves the right to modify any procedure that could damage the vessel. Customer and any personnel brought aboard by customer agree to follow all safety procedures and policies implemented by the captain.

### **3.3.6 Dive Safety**

The RMP requires only non-decompression SCUBA diving for bioaccumulation deployment, maintenance and retrieval. This section documents the safety procedures involved, responsibilities of the dive team members, equipment used and maintenance procedures, and emergency procedures associated with each of these types of diving.

AMS performs diving operations in a number of areas that are only accessible by water. The general procedure for accessing a dive site is to tie the vessel off to a fixed structure and attach a life ring directly to the structure prior to beginning dive operations. In all RMP diver operations, two divers and one dive tender are used; the two divers are tethered together. AMS uses underwater communication gear to allow the divers to communicate with each other and with the dive tender during the dive. General safety procedures use the following guidelines:

1. Boating Safety. A safety briefing is given by the skipper of the vessel prior to departure from the dock. This briefing shall include at a minimum the location of flotation devices, location of fire extinguishers, and emergency procedures. Enough information is given to allow the crew to communicate with emergency services, and operate the vessel in the event that the captain is incapacitated.
2. Equipment Storage. All equipment to be used as part of diving operations is properly stowed to minimize the possibility of movement during vessel transit.
3. Arrival at Dive Site. The dive tender assists the skipper in mooring the vessel to the fixed structure at each transplantation site. The tender shall also affix a life ring to the fixed structure, in case the vessel must depart from the structure while divers are in the water.
4. Pre-Dive Briefing. A briefing session is given by the lead diver or dive safety officer prior to beginning dive operations. The briefing includes such information as weather, currents, dive plan and emergency procedures.
5. Diving Safety. Each diver is responsible for performing a functional check of all dive equipment in the presence of the dive buddy or the tender. Dive operations are initiated given appropriate environmental conditions and appropriate health conditions of the divers. The dive operations can be terminated by either diver or the dive tender for environmental, diver health, equipment, or any other reasons. All AMS diving operations are conducted in accordance with procedures and requirements of the American Academy of Underwater Scientists (AAUS).
6. Underwater radio communications gear allowing communications between the divers and tender are used. This gear allows the divers to communicate even with the (commonly poor) visibility found in the San Francisco Estuary.
7. Debriefing. At the conclusion of each dive, the dive tender shall fill out the dive logs and the divers and tender discuss any issues of concern arising from the dive.

#### **3.3.6.1 Dive Team Member Responsibilities**

The responsibilities of the dive members are as follows:

1. Lead Diver - In addition to operations performed as part of the scientific investigations, the lead diver is responsible for planning the dive, discussing the plan with the diver and dive tender, and evaluating environmental conditions at each dive site.
2. Diver - In addition to tasks performed as part of the scientific investigations, the diver shall assist the lead diver in evaluation of the dive plan and environmental conditions. The diver also assists with mooring the vessel at the dive sites.
3. Dive Tender - The dive tender assists the vessel skipper with tying the vessel off to the fixed structure. The tender also participates in discussions of the dive plan and environmental conditions, assists the divers in preparing materials and equipment needed for dives, records information for dive logs, monitors dive communications, and assists divers returning to vessel.

#### **3.3.6.2 Dive Equipment Use and Maintenance**

Each diver is responsible for the safe functioning of their dive equipment. As such, each diver shall set-up their own gear and perform a functional check of all

equipment. Divers will make use of AMS equipment logs to record required inspection and maintenance of all equipment.

- Pre-Dive. Each diver shall be responsible for performing a functional check of all dive equipment in the presence of a buddy.
- Post-Dive. The divers shall follow-up on any equipment concerns that developed during the course of a dive prior to re-entering the water.

### **3.3.7 Dive Operations**

Owing to the relatively shallow dive depths and lengthy surface intervals spent in travel time between sites, the dive teams can complete several dives during the course of one day. Diving operations are developed with the conditions of the Estuary in mind and allow for safe diving in conditions of zero visibility, large wave swell and subsurface currents up to 1 knot. Dive operations have evolved over the course of the monitoring program to incorporate the highest degree of safety possible. Currently, the average dive operation at each site takes approximately 25 minutes. Table 17 presents a simplified outline of a bivalve bioaccumulation operation.

**Table 17. Dive operations task list.**

<i>Step</i>	<i>Task</i>	<i>Minutes/Task</i>
1	The vessel approaches piling and determines direction and strength of water current and prevailing wind.	3-4
2	Vessel backs up to piling, stern first, and a line is attached around the piling to the vessel's stern. The vessel's engine is turned off. A safety line with a flotation device on its end is attached around the piling and is placed in the water.  Should the vessel need to leave the piling while divers are under water, the safety line will remain attached to the piling.	3
3	Two divers enter the water from the vessel's stern and maneuver down the piling to the bottom.	1
4	Divers locate a ground line that is attached to the base of the piling. Occasionally, the ground-line is buried and must be dug out of the sediments by hand.	.5
5	Divers follow the ground line approximately 20-40 feet to where the line is secured to the bottom with a "screw-in type" ground anchor. Connected to the ground anchor is a 16 inch diameter plastic buoy attached to a 3-4 foot buoy line. Four bivalve bags are secured to the buoy line with cable ties.	.5
6	Divers perform their work on the bivalve bags by either installing, cleaning or removing bags. Divers check ground and buoy lines for secureness.	5-25
	Divers follow ground line back to base of piling.	.5
	Divers maneuver up the piling to water surface where vessel stern is still attached to piling. Divers board the vessel from the stern.	1
	The vessel's engines are started. The safety line is removed from the piling then the vessel's stern line is removed from the piling.	1

### **3.3.7.1 Dive Records**

During the cruise, the dive tender is responsible for maintaining dive logs for each dive. These logs record any comments pertaining to the dive, time and date, environmental conditions, and dive depth, time, and air usage.

At the conclusion of each cruise, each diver is responsible for completing an AMS dive log in accordance with AMS Dive Program regulations. These logs track all the dives completed by each diver over the course of a calendar year along with associated information listing the details of each dive.

### **3.3.7.2 Equipment**

Equipment needs will vary depending on the type of cruise operations (deployment, maintenance, retrieval). Table 18 presents an equipment list for bivalve deployment, maintenance and retrieval dives.

**Table 18. Equipment list for bivalve sampling cruise.**

<i>Quantity</i>	<i>Description</i>
6	Aluminum SCUBA tanks, 80 c.f.
3	Sets of personal dive gear (dry suit, regulator, gauges, fins, weight belt, gloves, knife, hood, buoyancy compensator)
3	AGA masks
3	Underwater OTS communication modules
36	AA batteries
1	Field medical kit
1	Life ring with 100 foot floating line
3	Tank straps for communications modules
1	Dive log book
1	Sample collection log book
5-7	Empty coolers, cleaned
1	Tool kit
3	Wire brushes (retrieval cruise only)
2	Boxes latex surgical gloves (retrieval cruise only)
3	Oyster knives (retrieval cruise only)
2	Rolls of aluminum foil, 100 ft. (retrieval cruise only)
65	Zip Loc bags, lg. (retrieval cruise only)
500	Cable ties, assorted size (retrieval cruise only)
2	Cable tie fastening tools (retrieval cruise only)

### **3.3.8 Dive Equipment Preparation**

1. Ensure that there are an appropriate number of complete AGA sets (including second stages, power supplies, and connectors) for the required dives.
2. Ensure power supplies are functioning properly. Replace old batteries.
3. Ensure each AGA set is in working order by attaching a working power supply, activating the push-to-talk switch, speaking into the microphone, and listening for reception.
4. Label each power supply with the date examined and the diver for which it is intended to be used on the cruise.
5. Clean and place equipment into Communication Gear Field Kit.
6. Each diver is responsible for performing a functional check of personal dive gear prior to each cruise.

### **3.3.9 Bioaccumulation Sample Handling**

In addition to the ship's log, a sample record must be maintained for each site. The sample record contains the following information:

1. Station name and code.
2. Species type.
3. Date of deployment / maintenance / retrieval.
4. Condition of bivalves.
5. Allocation of bivalves among various analyses.
6. Other remarks.

For the deployment cruise, the bivalves are stored in clean coolers on wet ice. Any accumulated water is allowed to drain off. For the retrieval cruise, once the bivalves are returned to the surface they are immediately placed into a cooler. The bivalves are processed either on the ship's back deck (if vessel is not underway) or within the ship's cabin or on the front deck (if the vessel is underway) to prevent contamination of samples by hydrocarbons in the vessel exhaust.

The initial processing of the bivalves is completed using clean, powder-free latex gloves. Dead bivalves are counted, recorded, and removed from the sample. Live bivalves are then allocated among analyses for organics, trace metals, and condition index (in descending order of priority). The numbers allocated to each of the analyses is then recorded on the sample logs.

The samples allocated to organics analysis are wrapped in aluminum foil, placed in a pre-labeled Ziploc™ bag, and placed in a cooler on dry ice. The samples allocated to trace metals are placed in two layers of Ziploc™ bags, one of them pre-labeled, and placed in a cooler on dry ice. Each labeled bag is marked with the date and number of bivalves.

The samples allocated for condition index do not require "laboratory clean" standards as there is no chemical analysis required. These bivalves are scraped to remove as many of the fouling organisms as is practical, rinsed, and dried. The bivalves are then secured with cable ties to prevent loss of tissue material, placed in a pre-labeled Ziploc™ bag, and stored on dry ice.

All samples are stored on the vessel for the duration of the cruise. Dry ice is replenished as needed to keep samples frozen.

The sample collection form, coupled with a chain of custody record and a laboratory analysis record, allows tracing of the complete history of a sample from time of collection to final entry of data to a computer database.

### **3.3.10 Bivalve Handling and Storage**

At the conclusion of the retrieval cruise, the samples are all transferred to AMS and placed in on-site freezers. The samples for trace metals analysis are shipped overnight on dry ice to GERG. The samples for organics are transferred by AMS personnel to UCSC for homogenization in preparation for laboratory analysis. The samples for condition index are maintained at AMS until they are processed. All shipped samples are shipped overnight in coolers with enough dry ice to keep samples frozen for at least two days.

### **3.3.11 Bivalve Chain of Custody**

Chains of custody (COC's) are completed each time control of samples is transferred from AMS to a receiving laboratory. For bioaccumulation samples, in addition to the standard shipping information, the following information is required:

1. Cruise number
2. RMP station name and code
3. Collection date
4. Species type
5. Analysis required
6. Other remarks

Completed COC's are either faxed to the receiving laboratory one day in advance of the shipment or hand-delivered with the samples. Receiving laboratories are requested to confirm delivery of samples by contacting AMS and by sending completed COC's to SFEI.

## 4 Appendix A. Figures

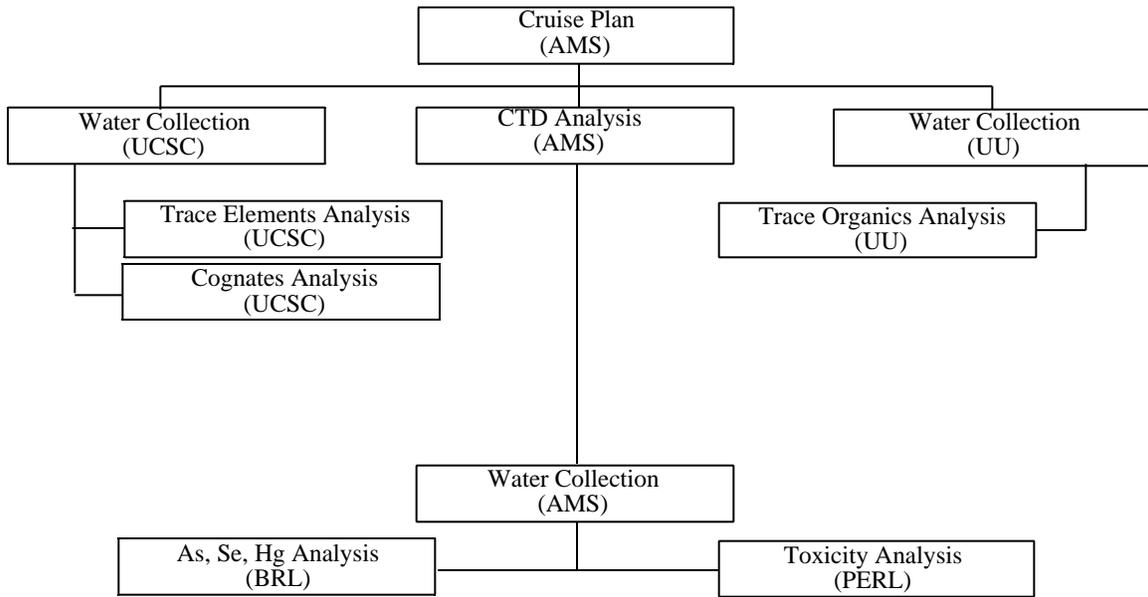


Figure 1. Organization chart for sample collection and analysis of water samples collected for the 1998 Regional Monitoring Program

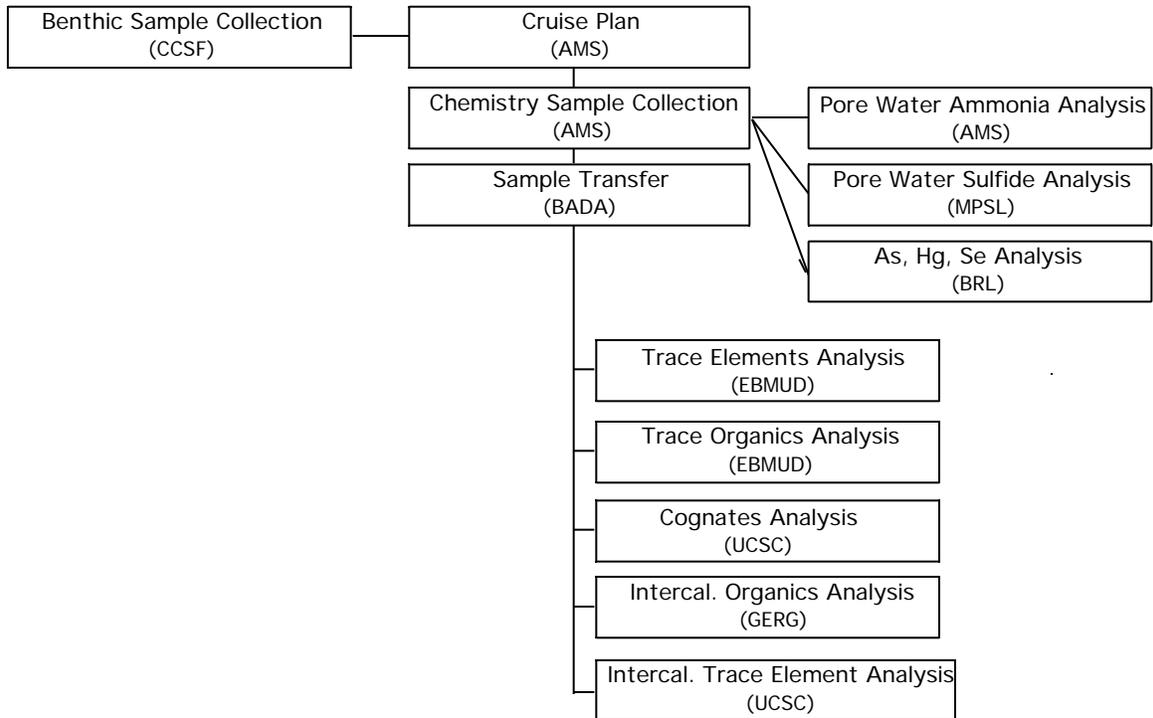


Figure 2. Organization chart for sample collection and analysis of sediment samples collected for the 1998 Regional Monitoring Program

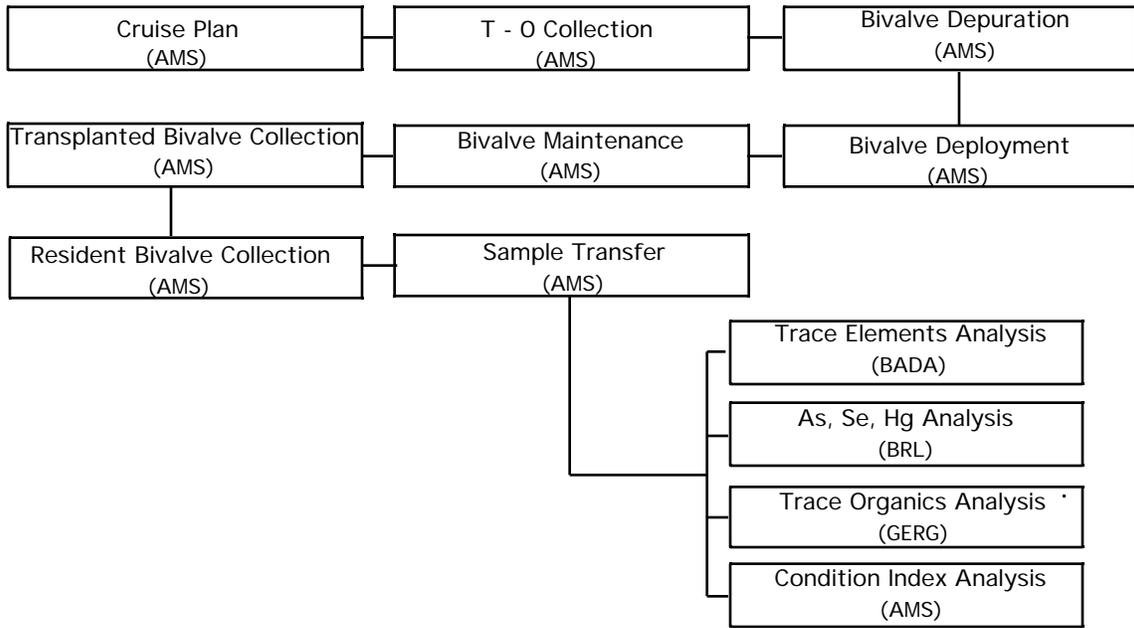


Figure 3. Organization chart for sample collection and analysis of tissue samples collected for the 1998 Regional Monitoring Program