

Data Collection Protocol

Tidal Marsh Benthic Community

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Introduction

Benthic infauna is primarily 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. They also serve as a main food resource for many intertidal fishes and birds (Goals Project 2000). They serve as important components of many ecological monitoring programs.

Benthic infauna can be classified as state indicators of the intertidal benthic community, and as pressure indicator of food resources for intertidal fishes and birds that feed on benthos.

Personnel

Any trained person can use this protocol. Training may require close supervision in the field to check on techniques and stratum selection.

Sampling Approach

Sampling Universe, Sites, Strata, and Sample Units

The sampling universe is defined by the distribution of unvegetated intertidal sediments in the San Francisco Estuary. A site is a wetland project or a patch of intertidal habitat. The primary sampling strata consist of the soft bottom substrates of intertidal flats, tidal marsh channels, and tidal marsh pannes

The sample unit consists of the benthos contained in 0.05 m² of sediment from 0 to 10 cm deep. A consistent sample unit volume is required to compare strata and sites, and to avoid introducing error due to species-area effects (Connor and McCoy, 1979). Because of this effect, any sub-samples used to compose a sample unit should be taken from contiguous locations, and be of the same volume. The sample unit volume 0.05 m² is recommended so that the results from wetland sampling will be consistent with other, recent San Francisco Bay studies, including the Regional Monitoring Program for Trace Substances (RMP), NOAA, and the Bay Area Clean Water Association (BACWA).

A stratified-random sampling approach will be used. For each sample site, a GIS is used to generate random points. The nearest points on the nearest small channel, large channel, and panne are designated as sample stations. Thus, there will be three stations for each of the three strata, for a total of three replicate sample units per stratum.

Benthic Sampling Procedure

The following procedure should be used to sample tidal marsh sediments for benthos where a vessel can access the larger channels and the Ponar grab sampler can be deployed from the vessel.

Ponar Grab in Large Channels

1. One sample unit is taken at each large channel station using a 0.05 m² Ponar grab sampler. The Ponar grab is lowered either by hand or with the aid of a winch (if available) from a boat at high tide. The grab is equipped with hinged stainless steel mesh lids with rubber flaps to allow flow-through of water during decent and retrieval to minimize disturbance of surface sediments and to trap organisms on the sediment surface.
2. A sample is determined to be acceptable if:
 - a. the grab jaws are completely closed
 - b. there is no evidence of washout through the mesh lids
 - c. sediment is evenly distributed in the grab
 - d. minimum disturbance of the sediment surface
 - e. minimum depth is 10 cm in clay/silt substrate, 5 cm in sandy substrates
3. After the core is accepted, it is placed in a rectangular plastic receptacle and taken to the sieving station.
4. The station identification, depth of water, depth of sediment penetration, sediment texture and color, and time of collection, weather conditions, replicate number, and any comments are recorded for future reference

Box Frame in Pannes and Small Channels

1. One sample unit is taken using a 0.05 m² box corer or other gear that is comparable to the Ponar grab. To be comparable the device must capture the sediment to a depth of 10 cm below an area of 0.05-m².
2. The corer is pressed into the substrate with evenly applied pressure (pressed by hand being careful to minimize the disturbance of the surface sediments) until the sediment level reaches the 10 cm depth mark.

3. A sample is determined to be acceptable if:
 - a. sediment is evenly distributed in the grab
 - b. minimum disturbance of the sediment surface
 - c. minimum depth is 10 cm in clay/silt substrate, 5 cm in sandy substrates
4. The station identification, depth of sediment penetration, sediment texture and color, and time of collection, weather conditions, replicate number, and any comments are recorded for future reference.

Glass Tube in Pannes or Small Channels

1. All samples are taken with a thick-walled, 1.5 m long glass tube, having an inside diameter of 10 cm. (note: 6.5 cores would be equivalent to one Ponar grab)
2. The tube is inserted to a depth of 10 cm. If the depth to resistance is less than 7 cm, then another place for sub-sampling is randomly selected for the stratum. As the glass tube is inserted, its top end is kept uncovered to prevent backpressure that could inhibit the sediment from entering the bottom of the tube.
3. The tube is extracted from the substrate by turning the tube in a twisting motion to break the connection between the substrate and the sediment in the tube. Before the tube is pulled from the substrate, a stopper is placed over the top.
4. The total length of the core in the tube is measured to the nearest 1 cm. Reading the length of the core will require cleaning the outside of the tube.
5. The station identification, depth of water, depth of sediment penetration, sediment texture and color, and time of collection, weather conditions, replicate number, and any comments are recorded for future reference.
5. The core is extruded from the tube into the rectangular plastic receptacle, and the inside of the tube is carefully rinsed into the receptacle so as to capture the entire contents.
6. Repeat this procedure to collect 7 cores (equivalent to one Ponar Grab) and proceed to the sieving station. Make sure the cores are collected right next to each other (in a effort to make the different sampling techniques as similar as possible).

All Strata

After the core is accepted, a scoop is used to collect the sample out of the corer and is placed in a rectangular plastic receptacle and taken to the sieving station. Care must be taken not to remove more sediment than is equivalent to the bottom of the corer (make it level with the bottom edge).

In very unconsolidated sediments it may be difficult to collect an exact volume as the substrate may try to fill the receptacle from the bottom (do the best you can).

Sample Sieving, Preparation and Processing

1. At the field wash station, the sample is gently washed through the nested 1 mm and 0.5 mm sieves using the battery operated water pump system.
2. The clean material retained in each screen is gently washed into separate, clearly labeled sample jars. Jars should have a small paper label on the inside and on the lid which specify: study, date, station ID, sieve size, jar number (e.g. 1 of 2, 2 of 2).
3. A relaxant (isotonic $MgCl_2$) is added to the sample for 15-30 minutes.
4. The relaxant is poured off (using a screened lid) prior to being fixed in 10% buffered formalin.
5. As a final step, two to three drops of stain (rose bengal solution) are added to the sample for ease of organism sorting.
6. Samples should be transported in a box that ensures that the glass jars will not break.
7. Within 3 days, samples should be rinsed in tap-water and transferred to 70% ETOH.
8. Samples are sorted into major taxonomic groups and then identified to lowest identifiable taxon by qualified taxonomists.

Interferences

Sample Loss or Damage - The potential for sample loss or damage is present at several steps during the collection and preservation process. Care must be taken to ensure: 1) sample loss does not occur by overfilling sample buckets when emptying the bottom grab; 2) damage to delicate invertebrates does not occur during the sieving process by setting water pressure too high or careless picking of organisms from the sieve screens; 3) sample loss does not occur by allowing clogged sieve screens to overflow; and 4) samples are properly relaxed, fixed, and preserved.

Formalin - The buffering capacity of the sodium borax in the formaldehyde decreases with time. If the pH of the buffered formaldehyde does not measure at least 8.2 prepare fresh fixative.

Timing of Sampling Effort

Sampling of large channels should be done from a boat using a Ponar grab at slack high tide. Samples from other strata can be taken at any tide stage.

Equipment and Materials

Sample Containers and Preservation

Sample Receptacle - Samples are emptied from the bottom grab into a large rectangular plastic tub.

Sample Jars - Sieved samples are washed into wide-mouth, glass, 1 qt canning jars equipped with dome lids and bands.

Anesthetic - Samples are relaxed in a solution of MgCl₂-6 Hydrate isotonic with seawater.

Fixative - Samples are fixed with pH buffered 10% formalin in ambient water.

Preservative - Samples can be preserved indefinitely in 70% ethyl alcohol in water. Tap water should be used to rinse the fixative from the samples within 48 to 72 hours of collection (samples should not remain in the fixative longer than 7 days).

Staining - Two or three drops of concentrated rose bengal solution are added to each sample at the time of fixation to aid in sample sorting.

Storage - Samples may be stored at room temperature. Sample jars are secured in plastic trays with dividers while on board the sampling vessel.

Research Vessel - For benthic sampling in the deeper channels a Zodiac or a Boston Whaler type vessels are appropriate. The vessel must be large enough to accommodate crew and equipment. The vessel must also be equipped with safety equipment.

Field Equipment

Bottom Grab - Use a Ponar grab (core area is 0.05 m² (500 cm²)) for finer sediments and any necessary equipment for proper use (e.g. cocking bar, weights, etc.). The grab can sit in a large plastic tub.

Small channel collection tube: A 1.5 m long plastic tube, having an inside diameter of 10 cm. A large spatula that covers the bottom of the tube, a stopper, and gloves.

Frame corer: A metal or plastic box (with the bottom cut out of it), having an inside area of 500 cm² (or about 22.5 cm x 22.5 cm), with scored markings at 10 cm from the bottom edge, and a large scoop (used to scoop out the sample from the box).
Sample Collection - A logbook contains the necessary data sheets for the cruise. Equipment used during sample collection includes a plastic ruler in centimeters to measure depth of

sediment collected in the grab, plastic scrapers (optional) for pushing sediment from the grab, a plastic tub for receiving sediment from the grab, quart canning jars and lids to contain sieved samples, preprinted labels for jar lids, preprinted waterproof labels for inside of jars, waterproof pens, pencils, and plastic trays with dividers for securing sample jars.

Sieving Equipment - Equipment for sieving benthic samples includes rectangular plastic tub equipped with a siphon hose for holding sieves and carrying sediment overboard, nested sets of 1.0 mm and 0.5 mm sieve screens for sieving sediment, fine forceps for picking organisms from sieve screens, stiff-bristle brushes for cleaning sieve screens between samples, canning funnels to aid in transferring samples from sieve screens to sample jars, two batteries to operate the water pump (2nd one is a back up), water hose assemblage attached to the pump to pump water from the channel for washing the sample, flow control fan-shape nozzle to allow for controlled washing of sediments through sieve screens and into sample jars (optional), plastic squirt bottles for rinsing sieve screens and canning funnels into sample jar, and tool box to store equipment.

Anesthetizing, Fixation and Staining Equipment - Equipment includes large plastic container for dispensing the anesthetizing and fixing reagents, jar-lid screens (0.25 mm mesh), one small squirt bottle for each reagent to wash the jar-lid screens, a small dropper bottle for the stain, safety glasses, nitrile gloves, and portable eyewash.

Personnel Equipment - Persons performing benthic sampling should wear foul weather gear, boots, and optionally gloves and life vest.

Reagents

Isotonic Magnesium Chloride - In the laboratory prepare MgCl₂-6 hydrate isotonic with sea water by dissolving 73 g in one liter of tap water. Prepare a volume sufficient to anesthetize all collected samples.

Buffered Formalin - In the laboratory buffer full strength formalin (37% formaldehyde) by adding approximately 114 g of sodium borate (Na₂B₄O₇) per gallon and mixing thoroughly. Test for pH of at least 8.2 with litmus paper. In the field prepare 10% formalin solution by mixing 1 part of the buffered formaldehyde with 9 parts sea water in a calibrated carboy.

Rose Bengal - In the laboratory prepare rose bengal solution by adding approximately 1/4 teaspoon of rose bengal powder to 100 mL tap water in a small dropper bottle.

Ethyl Alcohol - In the laboratory prepare 70% ethanol by mixing 7 parts 200 proof ethyl alcohol with 3 parts tap water. Use of undenatured ethanol requires a permit from the U.S. Department of Treasury, Bureau of Alcohol, Tobacco and Firearms.

Results and Analysis

Raw data consist of a list of identified taxa and abundances found in each sample for each sieve size. Identification of benthic assemblages at the sites sampled will be accomplished using ordination and classification analysis (Smith et al. 1988) and comparisons of the results with the assemblages reported by Thompson et al. (2000). Evaluation of relationships between benthos and abiotic variables will be conducted using principal components analysis and multiple regressions with SAS.

Costs

The estimated cost per sample unit is \$1,700.00-\$2,000.00

Presentation

The data are presented in a summarized graphical form as the average number of taxa and abundances (Figure 1). Other ways of presenting results are in tables (Table 1).

Figure 1. Example graphic of the average number of taxa and abundance (per 0.05m²) of benthic infauna sampled in the San Francisco Estuary (1994-1997).

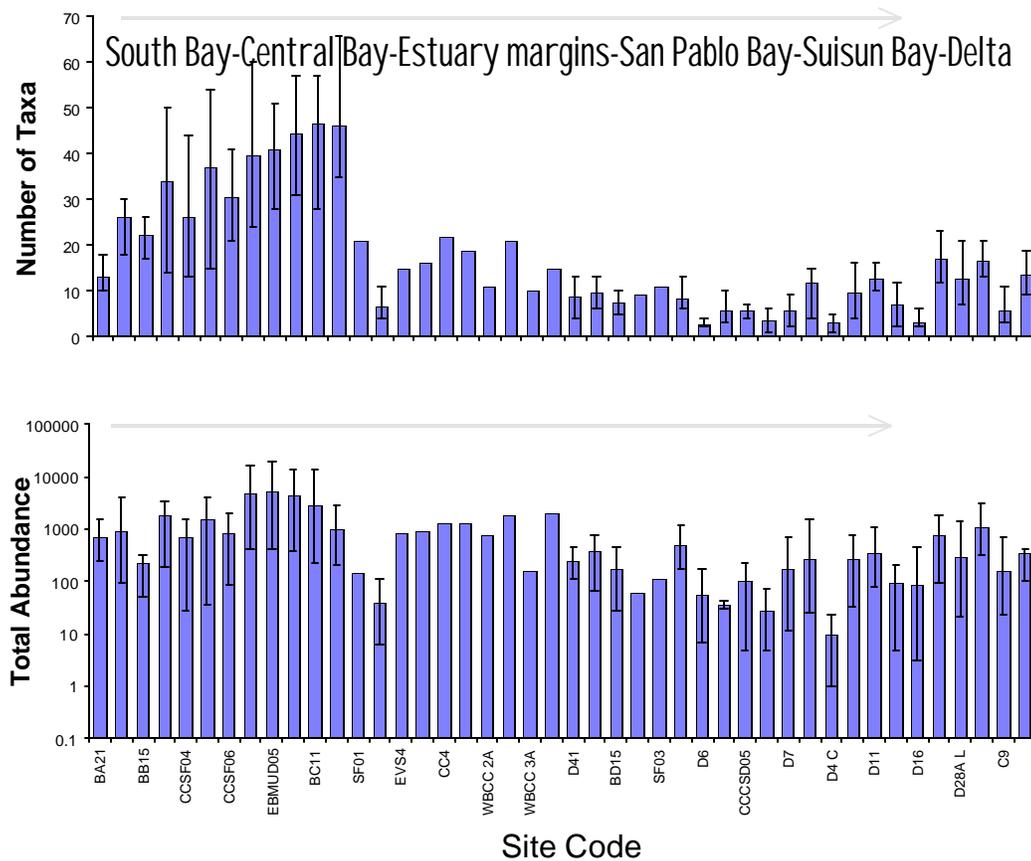


Table 1. Example table of the ranked correlation coefficients between number of taxa and total abundances and several abiotic variables in each sub-assembly, 1994 – 1996.

TOC = total organic carbon, mERMq = mean ERM quotient.

	Salinity	Fines	TOC	mERMq	
Central Bay marine muddy (n)	(60)	(60)	(60)	(59)	
No. Taxa	0.361**	-0.428**	-0.251	-0.320*	
Tot. Abundance		0.342**	-0.024	-0.071	-0.037
Main estuarine	(46)	(68)	(68)	(20)	
No. Taxa	0.343*	-0.094	0.057	-0.013	
Tot. Abundance		0.248	0.185	0.183	0.005
Estuarine transition	(70)	(71)	(71)	(20)	
No. Taxa	-0.015	0.503**	0.381**	0.048	
Tot. Abundance		0.241*	0.562**	0.513**	0.057
Fresh-brackish muddy	(167)	(192)	(192)	(0)	
No. Taxa	-0.263**	0.446**	0.485**	-	
Tot. Abundance		-0.130	0.424**	0.356**	-
Fresh-brackish sandy	(6)	(17)	(17)	(0)	
No. Taxa	0.415	0.050	0.420	-	
Tot. Abundance	0.065	0.022	0.190	-	

* $p < 0.05$

** $p < 0.01$

References Cited

- Connor, E.F., and E.D. McCoy. 1979. The statistics and biology of the species-area relationship. *American Naturalist* 113:791-833
- Goals Project. 2000. Bayland ecosystem species and community profiles: life histories and environmental requirements of key plants, fish and wildlife. Prepared by the San Francisco Bay Area Wetlands Ecosystem Goals Project. P.R., Olofson (ed). San Francisco Bay Regional Water Quality Control Board, Oakland, CA.