

Standard Operating Procedure

Great Salt Lake Water Quality Studies

Sampling of Benthic Zone

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The objective of Project 2A is twofold. First, we will develop methods for collecting the benthic periphyton/detrital mass that brine flies feed on, and the larval and pupal flies themselves. Secondly, these benthic samples and the overlying water taken in the production zones (water at 1-5 m depth) will be used for selenium analyses.

Sampling Sites

1. Bridger Bay on NW corner of Antelope Island near shorebird study area.
2. SW corner of Gilbert Bay near Kennecott outfall and additional shorebird study area.

Sampling Substrates and Depths

In each zone we will focus on sampling stromatolites, the principal habitat of brine flies, but we will also sample on sand and mud substrates.

1. Stromatolites (3 depths/zone): 1, 3, 5 m
2. Sand (1 depth/zone): 3-5 m
3. Mud (1 depth/zone): ca. 5 m

Sampling procedures

1. Water samples (lab preparations):
 - a. Bottles, 250 mL polyethylene:
 - Label (Note that the label should be covered with clear packaging tape to protect it.)
 - Wash with 1N hydrochloric acid in lab
 - Rinse 4 times with deionized water (DI), cap & place bottle in plastic bag
 - b. Sampling syringes, 60 mL (4/depth; Prepare 16 total)
 - wash in Liquinox
 - rinse 4 times with DI
 - fill with 1N hydrochloric acid and let sit for 30 minutes
 - eject acid and rinse 4 times with DI
 - place in new, clean plastic bags
 - c. Clean syringe filters
 - Soak 47-mm filter holder in 1-N hydrochloric acid for 30 minutes
 - rinse 4 times with DI

- place in plastic bag
 - in field, remove syringe filters, insert 47-mm, 0.45 μm membrane filters with acid-cleaned and DI-rinsed forceps. Fill syringe with 60 mL DI, attach filter holder and eject 60 mL of water through filter to rinse.
- d. Field sampling by SCUBA divers:
- First collect water samples at each site to minimize bottom stirring by divers.
 - Divers will take 4 syringes to each site in plastic bags.
 - Approximately 0.5 m above substrate rinse syringe 3 times by repeated plunges of plunger. Eject all rinse water. Move to undisturbed bottom.
 - Place syringe intake 1 cm above stromatolite, sand or mud and draw sample. Place filled syringe in bag. Move 1 m laterally and draw second sample. Repeat until 4 syringes are filled. Bring syringes to surface.
 - In boat, attach filter holder to sampling syringe and eject 40 mL of sample water through filter to rinse. Use this water to rinse the sample bottle. Shake vigorously and discard water from bottle. Fill bottle with remaining 20 mL. Attach 3 other syringes and filter all 60 mL water from each remaining syringe into bottle for a total sample of 200 mL.
 - Add 1% (2-mL HNO_3 (Ultrex grade)) to each bottle using acid-rinsed disposable pipette tip. Cap bottle and place in plastic bag then in cooler.
2. Stromatolites: Periphyton, larval, and pupal brine flies
- a. Containers:
- i. Periphyton. Rinse ten 500-mL plastic jars with 1-N HCl. Rinse 4 times with DI. Place label tape on each jar.
 - ii. Brine flies. Rinse 50 heavy-duty 1.5-gallon zip-lock bags with DI.
- b. Rinse two 500- μm screen sieves repeatedly with surface lake water to remove any sediments
- c. Brine flies will be sampled on hard stromatolite surfaces by SCUBA divers using a vacuum pump sampler similar to that of Voshell et al. (1992). The sampler consists of an inverted plastic canister with a port and glove attached to the side of the canister so that a diver can agitate the substrate. A hose attached to the bucket allows the sample to be brought to the surface with a hand-powered pump and to be collected in a net.
- i. *Site selection.* A weighted marker line will be dropped from each side of the boat. Divers will deploy down this line, bringing the canister with them. Sampling will commence on the nearest stromatolite surface large enough to accommodate the whole canister so that it covers only stromatolite and not other substrates.
 - ii. *Sampling at depth:* Scrub stromatolite surface with 3" diameter brush until "all" organic material, including flies, has been removed from stromatolite surface (ca. 1 minute). Release a float to advise boat observer to start pumping. Clear all initial water in hose (between the bucket and the pump) from the line, then pump a 12-L sample into a bucket or until pumped water is free of benthic organic material.

iii. *Sieve the sample* through a 500- μm sieve in boat. Next rinse sample into clean enamel pan. Assess how many pupae and larvae are present. We will need approximately 2000 of each life stage in order to get a 0.2-0.5 g (dry weight sample). If there are not enough brine flies or periphyton, advise divers to take a second sample at the same site.

iv. *Store samples* in labeled zip-lock bags and in cooler on ice (H_2O).

v. *Brine fly processing* in lab or on boat.

- place sample in acid-washed/DI-rinsed enamel pan
- Count and identify all larvae and pupae in sample. Only *Ephydra cinerea* are expected, but insure that no *E. hians* are present.
- Randomly pick ca. 2000 of each life stage (larvae, pupae) and place in acid-washed, DI-rinsed plastic scintillation vials for Se analyses. Rinse organisms 3 times with DI to remove salts. Place labeled vials with organisms in freezer.
- Place ca. 100 larvae (100 mg dry wt) in scintillation vial, rinse 3 times with DI and place in drying oven at 70° C for 24 hr for $^{13}\text{C}/^{15}\text{N}$ analyses. Grind and encapsulate for mass-spectrometer analyses.

vii. *Periphyton processing* in lab.

- Weigh entire sample of organic matter from the bucket.
- Homogenize sample and take sub-samples for:
 1. **Selenium.** If only a small amount of material is present rinse 3 times with DI and freeze for subsequent Se and AFDW analysis. If adequate material is present, preserve one as just described. The second sample can be treated to remove carbonates by placing sample in a 200-mL beaker and coving with ca. 160-mL, 1-N HCl to digest carbonates. Rinse 3 times with DI. Freeze in scintillation vial.
 2. **Chl a.** Take ca. 1 g of material, weigh it. Place in scintillation vial and freeze. Hold < 1 week. Cover with 15 mL, 95% ethanol. Extract overnight in dark. Measure chlorophyll fluorescence. Dilution may be required to stay on scale.
 3. **$^{13}\text{C}/^{15}\text{N}$.** Place ca. 5 g (mL) of material in 50-mL beaker. Add 40 mL, 1-N HCl. Leave for ca. 4 hr to allow carbonates to digest. Rinse resulting “pure” organic matter 3 times with DI. Examine with dissecting scope to insure that no carbonates are present. Dry at 70° C for 24 hr. Cool in desiccator. Grind and encapsulate samples.
 4. **Ash-free dry mass.** Rinse salts from approximately 5 g of organic material in tared crucible. Place in drying oven at 70° C for 24 hrs. Measure dry weight. Dry 1-2 hrs more and re-weigh to insure that all water has evaporated

from sample. Record weight. Combust in muffle furnace according to Standard Methods (use protocol for when carbonates present). Cool in desiccator and reweigh.

3. Sand & Mud: Periphyton/detritus, larval and pupal brine flies
 - a. Sampling device: Ponar Dredge
 - b. After locating sampling site, drop dredge from a distance of 1 m above the substrate
 - c. Bring dredge to surface and place in sieve
 - d. Carefully open dredge and remove ca. 50 mL of the substrate from the top 3 mm of the surface with a spatula. Place in whirlpac. Process as described above for:
 1. Selenium
 2. Chl a
 3. $^{15}\text{N}/^{13}\text{C}$
 4. Ash free dry mass
 - e. Open dredge completely into sieve and sieve contents over side of boat to remove brine flies. Place in zip-lock bag and in ice box.
 - f. Repeat until ca 1000 pupae and larvae are collected.
4. Adult flies - Collect with fine-meshed, loose-meshed net that can be grabbed and closed. Immerse net-end with flies in dry ice container with ca. 5 lbs dry ice. Place ca. 5 g. of frozen flies in labeled scintillation vial. Freeze.

Equipment and Supplies

1. 100 heavy-duty, 1.5-gallon zip-lock bags.
2. Butterfly net, fine, loose mesh
3. Large (1 ft³) soft foam dry ice container w/ 5 lbs dry ice
4. Bucket samplers
 - buckets, hose, pump (2)
 - scrub brushes (2)
 - 1-L squeeze bottles (2)
 - 500-mL sieves (2)
5. fine tweezer for picking brine flies (4); 1 clean set for changing filters
6. Scintillation vials (25/site)
7. Water sample bottles, 250 mL (10) EPA/top of cabinets
8. Buckets (size of bucket samplers) (2)
9. Large ice chest (?)
10. Block ice
11. Ponar dredge
 - rope (10-m)
12. 60-mL syringes
13. 47-mm filter cartridge
14. 47-mm, 0.45 membrane filters (20)
15. Nitric acid, Ultrex grade
16. enamel pan
17. 50 mL beakers
18. Ethanol
19. Mortar & pestle
20. Spatula

Boat

1. Table, 2 chairs
2. First-aid kit
3. Drinking/washing water 15 gallons in jugs
4. Dive ladder
5. 50' of 3/8" braided (not twisted) nylon anchor rope
6. 10', 3/8" nylon corner ropes for stern tie-downs of Poncho (10' each)
7. 2 marker lines for divers
8. 2 dive signal buoys for divers

SCUBA

1. wet or dry suits
2. weight belts (4)
3. tanks, filled (8)
4. mask, snorkels, fins
5. Regulators
6. Buoyancy compensators

7. depth gauge
8. dive flag
9. dive tables

Pump sampler

1. Hose
2. Sieve, 500 μm (buy)
3. Sampler

Ponar Dredge w/ rope

Eckman Dredge, rope, messenger

Periphyton (for chlorophyll & Se analyses; Organic carbon analysis)

1. Urine cups (20) with lids
2. Spatula for scraping surface

Adult brine flies

1. Butterfly net
2. Dry ice

Larval & pupal flies

1. 500-mL plastic jars 25