Bryozoan drift protocol

Incubation

1. Collect three bryozoan-covered kelp sporophytes (can by any kelp species, as long as you know which it is)
2. Cut five discs from each using the cookie cutter, making sure to include one with 0% and one with 100% cover (both sides); the remaining three somewhere in between
3. Randomly assign discs to 15 jars filled with seawater, keeping the 16th as a blank
4. Take pictures (with ruler and sample ID) of each side under an acrylic sheet with plenty of seawater to avoid air bubbles
5. Measure buoyant mass of each disc in a small plastic beaker to 0.001-g accuracy
6. Fill a tub with seawater and carefully take a triplicate of 50-mL, 0.45-µm filtered water samples with a syringe, avoiding a headspace in the tubes
7. Place a magnetic stir bar in each jar, close underwater avoiding bubbles, and randomly place on a magnetic stirrer under maximal lighting
8. Make note of time and leave for 24, 48 or 72 h
9. After the specified time interval, carefully open each jar in turn and first take a 50-mL, 0.45-µm filtered water sample, dispensing extra slowly to avoid equilibration
10. Keep all filtered water samples refrigerated as cold as possible in darkness (do not freeze!)
11. Re-weigh each disc as in step 5
12. Label 15 ziplock bags with the sample ID and freeze discs to be freeze-dried later
13. For each measurement round, measure PAR at each stirrer position
14. Measure jar volumes gravimetrically by filling the jar in a tub of MilliQ with and magentic stir bar, and then weighing the MilliQ to 0.001-g accuracy by decanting it into a tared beaker

pH and total alkalinity measurement

1. Calibrate the pH meter before every use using buffers and ensure a pH slope of >99%
2. The functionality of the titrator can further be tested using NaOH or $NaHCO_3$ standard solutions of known molarity made up in a 1 L volumetric flask with MilliQ
3. Take seawater samples from fridge and heat to room temperature using a water bath
4. Prepare 17 MilliQ-cleaned, labelled 50-mL volumetric flasks and carefully transfer 50 mL of each sample from tube to flasks using a small glass funnel and Pasteur pipette
5. Cap the volumetric flasks to minimise equilibration (the small headspace is ok)
6. Decant the solution from flask into titration vessel (plastic beaker) only just before titrating and take care not to agitate the water surface and cause bubbling
7. Record initial sample pH and temperature as well as total alkalinity
8. Carefully rinse the pH probe, burette and vessel with MilliQ between samples (do not physically dry the pH probe, merely absorb the drop at the tip with a Kimtech wipe)
9. Measure salinity of the remaining water in the tube using a reliable conductivity salinometer cross-validated with a digital refractometer

Data entry

1. The *seacarb* package in R needs a few variables to constrain the carbonate system: pH, total alkalinity, salinity and temperature
2. The titration procedure gives you total alkalinity and the initial pH and temperature will also be displayed but may need to be recorded manually, while salinity needs to be measured separately
3. Other useful titration data are the type of acid (sulfuric or hydrochloric), the molar acid concentration and the volume of acid added to reach the endpoint of the titration

4. To work with *seacarb*, total alkalinity will have to be converted to µM of alkalinity equivalents (typically ~2300); if the titrator's units are µM of $CaCO_3$, it's a simple multiplication by two because one CO_3^{2-} scavenges two H^+; in case of g of $CaCO_3$, the molar mass of $CaCO_3$ (100.0869 g mol^{-1}) must also be taken into account
5. Buoyant mass will be recorded before and after the incubation, while bryozoan cover needs to be measured via ImageJ later on as previously described in the other protocol
6. PAR at different stirrer positions and jar volumes are recorded in separate datafiles to the other data since we only want an average for these variables
7. The main datafile should look something like this:

ID	Round	Species	Individual	Mass	Cover	Time	pH	A_T	S	T
1_0	1	NA	NA	NA	NA	0	…	…	…	…
1_1	1	*Laminaria digitata*	1	…	0	48	…	…	…	…
1_2	1	*Laminaria digitata*	1	…	…	48	…	…	…	…
1_3	1	*Laminaria digitata*	1	…	…	48	…	…	…	…
1_4	1	*Laminaria digitata*	1	…	…	48	…	…	…	…
1_5	1	*Laminaria digitata*	1	…	1	48	…	…	…	…
1_6	1	*Laminaria digitata*	2	…	0	48	…	…	…	…
1_7	1	*Laminaria digitata*	2	…	…	48	…	…	…	…
1_8	1	*Laminaria digitata*	2	…	…	48	…	…	…	…
1_9	1	*Laminaria digitata*	2	…	…	48	…	…	…	…
1_10	1	*Laminaria digitata*	2	…	1	48	…	…	…	…
1_11	1	*Saccorhiza polyschides*	1	…	0	48	…	…	…	…
1_12	1	*Saccorhiza polyschides*	1	…	…	48	…	…	…	…
1_13	1	*Saccorhiza polyschides*	1	…	…	48	…	…	…	…
1_14	1	*Saccorhiza polyschides*	1	…	…	48	…	…	…	…
1_15	1	*Saccorhiza polyschides*	1	…	1	48	…	…	…	…
1_16	1	Blank	NA	NA	NA	48	…	…	…	…

A_T is total alkalinity in µM, S is salinity in ‰, T is temperature in °C