Squeal Daily Sampling Protocol– 2009

In the Field:

Field Equipment needed:

- 153μm zooplankton nets (x2)
- 147μm cups for zoop nets (x2)
- 2 squirt bottles w/ tap water—make sure they are full
- 4 zoop jars (4 oz.) for each lake (8 total) for gravimetric sample (A-D)
- 2 1L plastic bottles for chlorophyll
- ON FRIDAYS:
    - 2 extra zoop jars (12 total) for zoop chl sample
    - 2 4L plastic bottles for phycocyanin

→ Zooplankton:

- At the central sampling site in each lake (designated float), take a zoop tow
    - lower the net to appropriate depth (12m for Peter, 8m for Paul)
    - pull up the net at a constant rate of 3 seconds per meter
    - pour the sample into one of the 4 oz jars (start with A). Rinse the zoop cup several times with spray bottle and pour into jar
    - cap jar
- Move the boat a meter or two down the transect line between each zoop tow to avoid local depletion of zooplankton.
- ON FRIDAYS:
    - take 1 additional zoop tow and put sample into separate 4 oz jar marked "Fridays zoop chl"

→ Chlorophyll

- In each lake, with the 1L bottle, take a "glug" sample of water about a half meter deep. When taking a sample, make sure you don't collect any surface water.
- Don't leave the sample sit out for too long (chlorophyll is light and time sensitive). Between lakes, place the sample in the shade (not in the hot car).
- ON FRIDAYS:
    - collect a sample for phycocyanin by filling the 4L plastic bottle with surface water ("glug" just below the surface)

In the Lab:

Lab equipment needed:

- proweigh filters – 47mm (for zoop samples only)
- GF/F filters – 47mm
- ethanol – 75%
- film cans
- Folsom Splitter
- pre-filter – 2mm
- pre-filter – 80μm
- funnel
- Whirlpacks – 2oz

Right away, clean the zoop cups and zoop nets in the sink and hang them to dry

Filter for cholorophyll first (b/c it is light and time sensitive)

→ Chlorophyll:

- filter 200mL of water through a 47mm GF/F filter for each lake
    - Turn on vacuum line and make sure it is reading 200mmHg or less.
    - Place a filter grid-side down and then put on the magnetic filter holder.
    - Shake the sample bottle to resuspend anything that has settled.
    - Rinse the graduated cylinder 3 times with sample water.
    - Measure out 200mL of sample.
    - Pour sample into the filter holder and open valve.
    - After all of the water is gone, rinse filter holder with DI water.
    - Close calve, remove filter holder, and then carefully remove filter using forceps.
    - Place filter into a labeled film can and put it in the freezer.
    - Repeat above steps for replicate sample.
- when done, rinse graduated cylinder 3 times with DI and hang on drying rack to dry.
- EVERY FRIDAY:
    - Filter as much of the 4L phycocyanin samples through a 47mm GF/F filter as possible.
    - Fold filter in half and wrap in foil.
    - Label: phycocyanin, lake, date, and 47mm GF/F
    - Place in tuperware in freezer.

→ Gravimetric Zoops:

- Label each proweigh tin w/ lake, replicate (A-D), and date. Record filter weight on datasheet.
- Split each zoop sample using the Folsom splitter. If you do *not* split or if you quarter (etc) the sample, record it on the daily zoop data sheet; make sure to rinse the jar and lid of the jar into the splitter as well.
- Use half for zoop biomass sample and half for zoops in ethanol **(see directions below)**

- Pre-filter out the chaoborous using a 2mm mesh filter; carefully spray the filter to make sure the zooplankton go through. Either do this over a cup or over 80μm pre-filter to catch the zoops.
    - Paul zooplankton get stuck b/c they are so big—especially daphnia and holopedium
    - Peter zooplankton are tiny and stick to everything
- Please do a quick count of # of chaobs (big vs small) that went through into the zoop sample and record it on the data sheet in "comments".
- Filter each of the 4 gravimetric samples onto a separate proweigh filter, making sure to rinse the pre-filters and magnetic filter holder to wash zoops down onto the filter.
- Make sure to rinse splitter and pre-filters with DI between zoop samples.
- Once all zoop jars have been filtered, place filters in drying oven.
- Rinse zoop jars with DI water three times and place back into the coolers for the next day.
- Samples need to try for at least 2 days before they can be weighed.
- Each day, weigh the filters from 2 days before, which should now be dry, and record the total weight on the datasheet.
- EVERY FRIDAY:
    - After weighing the dried filters, save them all and ash them in the muffle furnace at 500°C for 4 hours. Then reweigh the filters and record on datasheet.
    - Filter the zooplankton chl sample the same way as the others (**but use 47mm GF/F instead of proweigh!**), but when done place it into a chlorophyll film can and put it in the freezer.

→ Zoops in Ethanol:

- for the other half of zoop sample from the splitter, filter out water (using 80μm pre-filter), and rinse into whirlpack using a funnel and 75% ethanol.
- NOTE: you do NOT need to remove chaoborous for these samples.
- You only need TWO zoops in ethanol samples. Dump the other two half zoop samples down the drain.