**222 and 221 Mega Sampling Plan: August 2013**

**BMC – 19 June 13**

MR comments in blue

**How to read the document:**

1. Shaded red areas are variables that have NOT been part of the every 3 week sampling this year or last summer.

2. I have added comments where I have questions or for things that I think need further discussion. If you are planning to skim this document, please skim to those areas.

3. I tried not to split up tables. So there may be pages with a lot of blank space…keep reading!

**Things that need to be decided ASAP:**

1. Are we sampling near, middle and far from a potential sampling point as described in the proposal? If so, we should probably take our samples in these locations in Aug as well to make our statistical analysis easier later on.

Great idea. Consider near, middle and far along a transect from drip point to outflow. A few sites (3-4) relatively close to the drip point, and more sparse further out. Consider spacing exponentially or something to concentrate sites at the drip point.

2. Where is the drip location? This is important for establishing the near, middle, and far sampling points described above. Mike Rennie has some suggestions for a drip point. I have included his description at the end. I suggest that we choose one this summer and drip a conservative tracer of some kind for a couple of days to see how the water moves. We can base our spatial samples off those data.

Also, great idea. Push hard for this one, as it will best inform our understanding of the mixing dynamics in the lake. Did you manage to get a look at the proposed drip site? Do you think it’s feasible? If so, we could easily do a transect from there to outflow for sampling.

3. What are the students doing for their dissertations? We need to make sure they collect the proper pre-drip data this summer. These projects need to be determined as soon as possible. **BEFORE we get to ELA this Aug.**

Nothing for me to say here.

4. It would be very helpful for me if we could meet in July (earlier is better) to finalize these plans…particularly the spatial sampling and drip point issue. I know that we are all spread out in July, but skype is a beautiful thing:) I would be happy to arrange the meeting if you send me your schedules.

Well done on getting this organized.

Pelagic Ag:

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Variable** | **collection** | **processing** | **preservation** | **analysis** | **Spatial Regime** | **Temporal Regime** |
| Total Ag (TAg) | Screen with 35 um mesh in field | 10 mls in 15 ml falcon tube | To final concentration of 4% nitric acid | Heat digest followed by ICPMS | Near  Middle  Far  Epi/Hypo | Every 3 weeks |
| Dissolved Ag (DAg) | Screen with 35 um mesh in field | Filter 10 mls with 0.2 polycarb | Filtrate to final concentration of 4% nitric acid. In 15 ml falcon tube | Heat digest followed by ICPMS | Near  Middle  Far  Epi/Hypo | Every 3 weeks |
| Seston Ag | Screen with 35 um mesh in field | Filter 300 mls with 0.8 poly carb | Filter in 5 mls of 4% nitric acid in a 15 ml falcon tube | Heat digest followed by ICPMS | Near  Middle  Far  Epi/Hypo | Every 3 weeks |
| BP Ag | Screen with 35 um mesh in field | Filter 300 mls with 1.2 ml poly carb followed by a 0.8um poly carb | 0.8 filter in 5 mls of 4% nitric acid in a 15 ml falcon tube | Heat digest followed by ICPMS | Near  Middle  Far  Epi/Hypo | Every 3 weeks |
| 1.2-0.2 fraction | Screen with 35 um mesh in field | Filter 100 mls with 1.2 polycarb followed by a 0.2 um polycarb | 0.2 filter in 5 mls of 4% nitric acid in a 15 ml falcon tube | Heat digest followed by ICPMS | Near  Middle  Far  Epi/Hypo | Every 3 weeks |
| 35-1.2 fraction | Screen with 35 um mesh in field | Filter 500 mls with a 1.2 polycarb | Filter in 5 mls of 4% nitric acid in a 15 ml falcon tube | Heat digest followed by ICPMS | Near  Middle  Far  Epi/Hypo | Every 3 weeks |
| Settling Ag | Sediments from traps | Filter 100 ml with 0.8 um polycarb | Filter in 5 mls of 4% nitric acid in a 15 ml falcon tube | Heat digest followed by ICPMS | Center Buoy | Every month |

Zooplankton:

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Variable** | **collection** | **processing** | **preservation** | **analysis** | **Spatial Regime** | **Temporal regime** |
| >80 um fraction abundance | 3 tows from 4m. 80 um net | Split sample | ½ sample preserved with sugar formalin (20 mls to 100ml sample) | Count and ID with scope | Near  Middle  Far | Every 3 weeks |
| >80 um fraction CNP | 3 tows from 4m. 80 um net | Filter onto 80um mesh. Scrape off biomass | Dry biomass in weigh boats | Elementar and P digestion followed by spec | Near  Middle  Far | Every 3 weeks |
| >80 um fraction Ag | 3 tows from 4m. 80 um net | Filter onto 80 um mesh. Scrape off biomass | Dry biomass in weigh boats | Acid and heat digestion followed by ICPMS. | Near  Middle  Far | Every 3 weeks |
| >80 um fraction 15N/13C | 3 tows from 4m. 80 um net | Filter onto 80 um mesh. Scrape off biomass | Dry biomass in weigh boats | Mass spec at Water Quality Center | Near  Middle  Far | Once in Aug |
| >20 um fraction abundance  (rotifers) | 3 tows from 4m. 20 um net | Split sample | ½ sample preserved with sugar formalin (20 mls to 100ml sample) | Count and ID with scope | Near  Middle  Far | Every 3 weeks |
| >20um fraction CNP | 3 tows from 4m. 20 um net | Filter onto 20 um mesh. Scrape off biomass | Dry biomass in weigh boats | Elementar and P digestion followed by spec | Near  Middle  Far | Every 3 weeks |
| >20 um fraction Ag | 3 tows from 4m. 20 um net | Filter onto 20 um mesh. Scrape off biomass | Dry biomass in weigh boats | Acid and heat digestion followed by ICPMS. | Near  Middle  Far | Every 3 weeks |
| >20 um fraction 15N/13C | 3 tows from 4m. 20 um net | Filter onto 20 um mesh. Scrape off biomass | Dry biomass in weigh boats | Mass spec at Water Quality Center | Near  Middle  Far | Once in Aug |
| >150 um fraction abundance (pred zoops) | 3 tows from 4m. 150 um net | Split sample | ½ sample preserved with sugar formalin (20 mls to 100ml sample) | Count and ID with scope | Near  Middle  Far | Twice in Aug |
| >150 um fraction CNP | 3 tows from 4m. Use 150 um mesh net | Filter onto 150 um mesh. Scrape off biomass | Dry biomass in weigh boats | Elementar and P digestion followed by spec | Near  Middle  Far | Twice in Aug |
| >150um fraction Ag | 3 tows from 4m. Use 150 um mesh net | Filter onto 150 um mesh. Scrape off biomass | Dry biomass in weigh boats | Acid and heat digestion followed by ICPMS. Send some to CO? | Near  Middle  Far | Twice in Aug |
| >150 um fraction 15N/13C nat abundance | 3 tows from 4m. Use 150 um mesh net | Filter entire sample onto 150 um mesh. Scrape off biomass | Dry biomass in weigh boats | Mass spec at Water Quality Center | Near  Middle  Far | Once in Aug |

Pelagic nutrients: Sample from epilimnion, metalimnion, and hypolimnion

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Variable** | **collection** | **processing** | **preservation** | **analysis** | **Spatial regime** | **Temporal Regime** |
| TDN/DOC | Screen with 35 um mesh in field | Pre filter with GF/F. Then filter with 0.2 polycarb | Fridge in acid washed amber glass bottles | Andrew does this at Trent | Near  Middle  Far  Epi/Hypo | Every 3 weeks |
| NO3/NO2 | Screen with 35 um mesh in field | Pre filter with GF/F. Then filter with 0.2 polycarb | Freeze in acid washed plastic 125ml bottles | Trent | Near  Middle  Far  Epi/Hypo | Every 3 weeks |
| NH4/TDP | Screen with 35 um mesh in field | Pre filter with GF/F. Then filter with 0.2 polycarb | Freeze in acid washed plastic 125ml bottles | Trent | Near  Middle  Far  Epi/Hypo | Every 3 weeks |
| TP | Screen with 35 um mesh in field | nothing | Freeze in acid washed plastic 125ml bottles | Trent | Near  Middle  Far  Epi/Hypo | Every 3 weeks |
| anions | Screen with 35 um mesh in field | Pre filter with GF/F. Then filter with 0.2 polycarb | Freeze in acid washed plastic 125ml bottles | IC? | Near  Middle  Far  Epi/Hypo | Twice in Aug |
| cations | Screen with 35 um mesh in field | Pre filter with GF/F. Then filter with 0.2 polycarb | Freeze in acid washed plastic 125ml bottles | ? | Near  Middle  Far  Epi/Hypo | Twice in Aug |
| DIC | Screen with 35 um mesh in field |  |  |  | Near  Middle  Far  Epi/Hypo | Twice in Aug |
| pH | Screen with 35 um mesh in field | Measure with meter at ELA |  |  | Near  Middle  Far  Epi/Hypo | Twice in Aug |
| 15N Natural abundance | Screen with 35 um mesh in field | Filter with 0.2 um (pre filtering may be needed) | Freeze in 1L plastic bottles | Digest and diffuse. Mass spec | Near  Middle  Far  Epi/Hypo | Once in Aug |
| 13C Natural abundance | Screen with 35 um mesh in field | Filter with 0.2 um (pre filtering may be needed) | Fridge in 1L amber glass bottles | Evaporate. Mass spec | Near  Middle  Far  Epi/Hypo | Once in Aug |

Pelagic standing stocks

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Variable** | **collection** | **processing** | **preservation** | **analysis** | **Spatial regime** | **Temporal regime** |
| Seston  chla | Screen with 35 um mesh in field | Filter with GF/F | Freeze in Al foil | Ethanol extraction followed by florometer | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| Pigments | Screen with 35 um mesh in field | Filter with GF/F | Freeze in Al foil | Ethanol extraction followed by HPLC | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| Seston CN | Screen with 35 um mesh in field | Filter with ashed GF/F | Dry in oven | Elemantar | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| Seston P | Screen with 35 um mesh in field | Filter with ashed GF/F | Dry in oven | Extraction followed by spec | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| BP chla | Screen with 35 um mesh in field | 1.2 um polycarb then GF/F | Freeze in Al foil | Ethanol extraction followed by florometer | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| BP CN | Screen with 35 um mesh in field | 1.2 um polycarb, ashed GF/F | Dry in oven | Elemantar | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| BP P | Screen with 35 um mesh in field | 1.2 um polycarb, ashed GF/F | Dry in oven | Extraction followed by spec | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| BP abund. | Screen with 35 um mesh in field | Screen 4 ml with 20 um mesh | Preserve with formaldehyde to 1%. Fridge until flash frozen. | Flow cytometer | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| Viral abund. | Screen with 35 um mesh in field | Screen 4 ml with 20 um mesh | Preserve with formaldehyde to 1%. Fridge until flash frozen. | Some kind of filtering followed by flow cytometer | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| Algal abund. | Screen with 35 um mesh in field | nothing | Preserve with lugols solution in clear glass bottles | Count and ID with scope | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| BP DNA | Screen with 35 um mesh in field | 1.2 um polycarb, then 0.2 polycarb | Roll filter and place in DNAase free tubes. Freeze | China? | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| BP resp. | Screen with 35 um mesh in field | CTC assay | Fridge until flash frozen | Flow cytometer | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| BP prod. | Screen with 35 um mesh in field | 3H labeled leucine assay | In scint vials | Scintillation counter | Near, Middle,  Far  Epi/Hypo | Every 3 weeks |
| Seston 15N/13C | Screen with 35 um mesh in field | filter with ashed GF/F | Dry in oven | Mass spec | Near, Middle,  Far  Epi/Hypo | Once in Aug |
| BP 15N/13C | Screen with 35 um mesh in field | 1.2 um polycarb, then ashed GF/F | Dry in oven | Mass spec | Near, Middle,  Far  Epi/Hypo | Once in Aug |

Littoral Sampling

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Variable** | **collection** | **processing** | **preservation** | **analysis** | **Spatial regime** | **Temporal regime** |
| Periphyton ID | Scrape tiles and save slurry | 40 ml of slurry | Preserve with lugols | Count and ID with scope | 5 locations (at least 3 tiles at each location) | Once in Aug |
| Periphyton CN | Scrape tiles and save slurry | Filter 10 mls slurry on ashed GFF | Dry in oven | Elementar | 5 locations (at least 3 tiles at each location) | Once in Aug |
| Periphyton P | Scrape tiles and save slurry | Filter 10 mls slurry on ashed GFF | Dry in oven | Digestion then spec | 5 locations (at least 3 tiles at each location) | Once in Aug |
| Periphyton chla | Scrape tiles and save slurry | Filter 10 mls on GFF | freeze | Ethanol extraction then flourometer | 5 locations (at least 3 tiles at each location) | Once in Aug |
| Periphyton 15N/13C nat abundance | Scrape tiles and save slurry | Filter 10 mls slurry on ashed GFF | Dry in oven | Mass spec | 5 locations (at least 3 tiles at each location) | Once in Aug |
| Periphyton Ag | Scrape tiles and save slurry | Filter 10mls slurry on 0.8 um polycarb | Filter in 5ml 4% nitric acid | Heat digestion followed by ICPMS | 5 locations (at least 3 tiles at each location) | Once in Aug |
| Macrophyte Ag | Collect all above ground tissue in grid | Dry in paper bags. Grind dry material. | Save dried | Some digestion followed by ICPMS | 5 locations (at least 3 grids at each location) | Once in Aug |
| Macroinvertebrate diversity – rock bags | Collect rock bags in ziplocks | Remove all macroinverts | Preserve in 80% ethanol | ID to genera and FFG using scope | 5 locations (three bags at each location) | Once in Aug |
| Macroinvertebrate Ag | Collect rock bags in ziplocks | Remove all macroinverts | Save individuals from representative taxa (FFG) and dry | Some digestion followed by ICPMS (or CO?) | 5 locations (three bags at each location) | Once in Aug |
| Macroinvertebrate diversity - OBBN | Kick sweeps |  | Preserve in 80% ethanol | ID to genera and FFG using scope | 5 locations following OBBN | Once in Aug |
| Sediment Ag | Cores Ekman grabs | dry | Save dried | Some digestion followed by ICPMS | Near  Middle  Far | Once in Aug |
| Sediment invert community | Cores Ekman grabs |  | Preserve in ethanol | ID to genera and FFG using scope | Near  Middle  Far | Once in Aug |
| Sediment bacterial community | Cores Ekman grabs | Freeze? |  | Extract DNA and DGGE or tRFLP | Near  Middle  Far | Once in Aug |

**Other experiments/sampling:**

Potential nitrification in sediments

Potential denitrification in sediments

Sediment enzyme activity

Organic matter decomposition

Metabolism using benthic chambers (ask Scott Higgins)

NDS to evaluate N and P limitation

Daily GPP, NEP, R (ask Scott Higgins)

**Equipment needed:**

Vacuum pump

150 um mesh zooplankton net

pH meter

wind meter

**Possible drip spots from Mike Rennie:**

If you head out along the west side of the lake from the landing (along the shore to the left- careful for that rock right off the launch), if you continue along, you'll find a small number "13" sign on a tree. It's right next to a gently sloping rock. THere's not a ton of room there presently, but I was thinking if we could get in with a chainsaw to take out a few dead trees (and live ones if necessary) it might make for a good working spot on solid ground. The other advantage to this spot is that it's pretty much opposite the side of the lake from the outflow, and would provide a decent gradient for nanosilver dispersal. I still want to have a look at a bath. map to get a better handle on what the depth gradient there is like. Might even be worth jumping in with a snorkel to see what the area below there is like.

The other potential spot I was looking at is essentially "around the corner" (around the point, more accurately) from where we set up to do our sampling. It would take some sussing out to find a decent enough spot that's relatively open, and not too mushy. The other concern with that area is that it is pretty close to the outflow, and depending on flow may result in only part of the lake receiving exposure (though I don't know enough about the flow rates to know whether that's a concern-most everyone here seems to think there won't be a gradient at all given that these small lakes mix so quickly).

My concern with other spots on the lake that are on soft ground is that it'll just become a huge muddy mess in no time, unless we consider building a large platform/deck on shore to work from.