1. Get moist towels (from home?)
2. Set out the jars with their tops off
3. Get the second sieve from the teaching lab so I don’t have to clean them in between. Also grab funnel holders and 50mL holders.
4. Create log sheet for extraneous analyses (masses, wet mass/dry mass)
5. Check septa for standard curve are in good enough shape
6. Get out the needles and set up the gas station
7. Test KOH with EC to find low enough mass for sufficiently precise value
8. Calculate the mass of wet soil that should be added to the KCl, MB, and Mehlich III extractions.
9. Calculate the mass of old soil that should be sieved, leaving the remaining in the incubation room.
10. Decide on the mass of wet soil to add to each jar (remembering some water loss throughout may result in insufficient soil at the end)
11. Decide on CO2 curve points, based on expectations (aim high for first one).
12. Label the CO2 curve jars.
13. Make sure the MB flushing set is working, and set up its station.
14. Make sure there is a 1000 pipette set to 500mL for chloroform.
15. Find the 5mL pipette for adding water to bottom of jars.
16. Check the volume of KOH needed for one filling (<0.1M).
17. Set up EC station with DIW, towels, etc.
18. Find tops for the rolling to mix BC + soil
19. Create the area where the filling, etc., will take place, with BC, 50mL jars, and vials.
20. Add 5mL CO2-free water to each jar, including STD curve jars.
21. Get soils from incubation room
22. Sieve both soils <2mm + cover with moist towel (like bread)
23. Initiate the microbial biomass extraction (this could actually be first, since it takes 4.5 hours of shaking).
24. Add 10g soil to each jar, except for the blanks.
25. Add 0.5mL Et-OH-free CHCl3 to the **+C** jars, and nothing to the **NO** jars, which all already have 40mL 0.05 M K2SO4.
26. Seal jars tightly, and put on shaker at 120 rpm for 4.5 hours (record time).
27. Bubble (the CHCl3) treatments with lab air for 15min (use fresh needles!).
28. Filter through (Whatman No. 1 or maybe 42) into glass jars.
29. Store extracts in fridge.
30. Prepare volumetric flask of KOH with CO2-free water. Mix very well.
31. Immediately take EC in flask + record value.
32. Pour flask into 1L (challenge - insufficient to hold total sample… fill 2 jars and don’t mix within same tray should be okay, so long as all mixed at one point).
33. Attach pump and flush out ~20x 15mL.
34. Fill vials in 8 STD curve jars, sealing each immediately after delivery. These can be later injected with CO2 from the tank.
35. Weigh out soils for tray 1 (4 young and 4 old) into bottles to .05 precision.
36. Add corresponding BC to each.
37. Roll bottles to mix, and then tap them down gently to settle soil uniformly. Be sure to roll the non-BC treatment as much as the BC treatments.
38. Place all bottles in Mason jars.
39. Add 15mL KOH to each vial, sealing the Mason jar immediately after. Add in a random order. Add the blank jar in the middle (5th). After adding and sealing all jars, re-check every jar to ensure they are tight. Record the time. At this point, may want to bring down to incubation room, depending on timing (?).
40. Set aside a little soil into the collecting jars for N, P, TEA, [MB], and moisture analyses.
41. Repeat previous 5 steps for each of the 8 trays.
42. Inject the correct volume of CO2 into each standard curve jar.
43. Weigh out 1.0g BC and add to corresponding 2M KCl vials (2 reps per BC)
44. Add Xg soil to corresponding vials (3 reps per soil).
45. Put on shaker at 120 rpm for X hours (record the time).
46. Filter through Whatman No. 42 (pre-rinsed with 2M KCl) into plastic tubes (for freezing?). May need to add extra for BC to get through filter.
47. Freeze the samples immediately
48. Weigh out 1.0g BC and add to corresponding Mehlich III vials (2 reps per BC)
49. Add Xg soil to corresponding vials (3 reps per soil).
50. Put on shaker at 120 rpm for 15 minutes (record the time).
51. Filter through Whatman No. 42 into glass jars. May need to add extra for BC to get through filter.
52. Store the extracts in the fridge.
53. Now should have all samples prepped, standard curve ready to be measured the next day, and pre-analyses ready to be submitted.
54. Label all the 50mL tubes with their correct ID number.
55. Distribute out 5mL of BaCl2 into each 50mL tube.