**Fractionation experiment optimization**

**General scheme**

1. **Add SPT to soil, shake lightly, centrifuge, filter, collect fPOM (wash filter and dry or weigh filter before/after and submit whole filter).**
2. **Add SPT to soil, sonicate/shake with glass beads, centrifuge, filter, collect oPOM (wash filter and dry or weigh filter before/after and submit whole filter). If beads need to remove.**
3. **Rinse, centrifuge with DW several times.**
4. **Shake with hexametaphosphate, sieve, collect silt+clay (dry in tared pan).**

**Question**

1. Glass beads or sonication.
2. Sample size – did 2 g. Can increase if not enough POM
3. Collection of POM (wash or submit filter)

**Answers**

1. With glass beads I can shake large batches overnight and avoid sample by sample sonication. Need to test amount of oPOM collected. If the same than use glass beads.
2. Washing filter sample means lost material and weighing, drying, and scraping many pans. Submitting filter means small sample size, weighing accurately and rolling filter to submit.
3. More soil means more SPT, larger vials etc. Less soil could mean not having enough POM to measure and submit. Question 3 is rolled up into 2.

**Experiments**

Dry 100g of soil to 60C. Dry 100g of soil and 2 g of litter at 60C.

SPT in small squirt bottle

Na-hexametaphosphate 0.5%

Question 1:

1. 2 g soil (+40 mg litter), 10 mL SPT. Disperse. Shake for 5 minutes, centrifuge (2500 g 20 min), float, filter thru something easy, measure fPOM, add back SPT into vials.
2. Add several glass beads to BEAD vials and shake overnight (12 hrs? 16 hrs?).

or

1. Sonicate at 450 J/mL in specimen cup (?) use squirt bottle to transfer back to vials.
2. Allow to float. Centrifuge (30 min 2500 g), tare weighing boat. Filter thru something easy, collect, dry at 60C and weigh.
3. Compare amount of oPOM collected between glass beads and sonication.
4. Rinse all vials with DW and centrifuge repeatedly.
5. Add (back glass beads and) HMP. Shake overnight. Sieve thru 0.53 um.
6. Collect sand (and POM?) and silt+clay in tared pans. Dry at 60 C.
7. Due LOI on sand. How much POM is left.
8. Compare weight of sand, silt+clay, and POM between sonication and glass beads.

Number of samples – 12

Treatment - glass beads/sonication, litter or no litter.

3 vials beads+litter

3 vials sonication+litter

3 vials beads+no litter

3 vials sonication+litter

Question 2:

1. 3 g soil (+litter), 15 mL SPT 1.7 g/cm3. Shake 30 times, float, centrifuge (2500 g 30 min).
2. Filter through tared 25 mm filters, wash with a lot of DW and place on tared weighing boats. Dry to 60C. Weigh and calculate amount of fPOM. Roll whole or sliced filters into EA capsules.

or

1. Filter through tared 25 mm filters, wash with a lot of DW, wash fPOM into tared pans/weighing boats. Dry to 60C. Weigh and calculate amount of fPOM. Scrape and weigh fPOM in to EA capsules.
2. Shake all soils with several glass beads overnight. Allow to float. Centrifuge (2500 g 30 min).
3. Filter according to (2) or (3).
4. Compare amount of fPOM and oPOM. Compare difficulty of preparing EA capsules, and drying samples. Decide on sample size (does 3 g block the filter?

Number of samples – 12. Soil 36 g. SPT 200 mL.

Treatment – litter or no litter, wash off or submit all filter.

3 vials litter-wash off

3 vials litter-submit all

3 vials no litter-wash off

3 vials no litter-submit all.