**Biogenic Si Analysis**

**In the Lab:**

1. If tube blanks did not get prepared during the cruise, use clean (HF rinsed) 15ml teflon centrifuge tubes, place them in the drying oven overnight so they've been heated the same as the tubes with samples. Treat blanks as you do all other vials with filters.

2. Cover filter with 4ml of 0.2N NaOH. Cap and vortex. Be sure filter remains submerged. Loosen caps but be sure the top of the tube is completely covered.

3. Place in water bath at **95°C** for:
   - RoMP samples = 1 hr
   - BATS samples = 2hr
   - JGOFS = 40 min
   - Plumes and Blooms = 30 min
   - LTER = 40 min
   - EqPac = 45 min
   - HOT = 45 min
   - IrnBru = 45min

4. Cool in ice water bath immediately. Samples should be cool to the touch before adding acid.

5. Add 1.0 ml of 1 N HCl and vortex after each addition. This neutralizes the NaOH and along with the cooling stops the digestion. Work quickly to minimize the time difference between the first and last sample.

6. With a Teflon rod or long forceps, crunch the filter into bottom of tube. Rinse the rod/forceps with Nanopure between samples. Scrunching the filter makes it easier to withdraw your sample later, but you will need to remove the filter eventually so be sure it’s not jammed into the bottom of the tube.

7. Centrifuge for 10 min at setting 6 (desktop clinical centrifuge) or program 4 (Thermo 20 tube centrifuge) to drive the lithogenic Si particles to the bottom of the tube.

8. Withdraw 4ml of the 5ml in the Teflon tube and place in 50ml PP (polypropylene) tube – be sure to take the sample from the top of the liquid, don't push the pipette tip down into the filter. Add 6ml of Nanopure to the PP tube for a total of 10ml. This 10ml will be used for dSi analysis – the dilution factor for this sample will be 5/4=1.25.

9. Follow same protocol as for Dissolved Si Analysis - 4 ml of the Acid/Moly reagent, wait 10 minutes to form silicomolybdic acid, add 6 ml reducing reagent. Make extra reagent blank if you think you might need to dilute!

10. Tube blanks correct for signal generated by the effect of the NaOH digestion on the tube. Filter blanks correct for signal generated by the effect of the digestions on the filter. A signal from the tube blank <90%T for 1cm cell, <80% for 10cm cell is high and may indicate a bad tube. The filter blank incorporates the tube blank, the signal from the filter blank is subtracted from the signal on all samples.