Carbonate Sample Preparation Procedures

Overview

This method is designed to isolate endogenic carbonates from lake sediment by removing larger biogenic carbonate, and as much organic matter as possible. The best place to perform this procedure is in SRCC 301 and 416 in one of the large basin sinks with surgical tube attached to the faucet. This procedure may be used as a preparation step when performing stable isotope, XRD, trace element chemistry, SEM, or other analyses.

Procedure

Sample

a. Follow **"Sampling Protocol"** and store sample in a 15 or 50 mL plastic Falcon Tube with appropriate label.

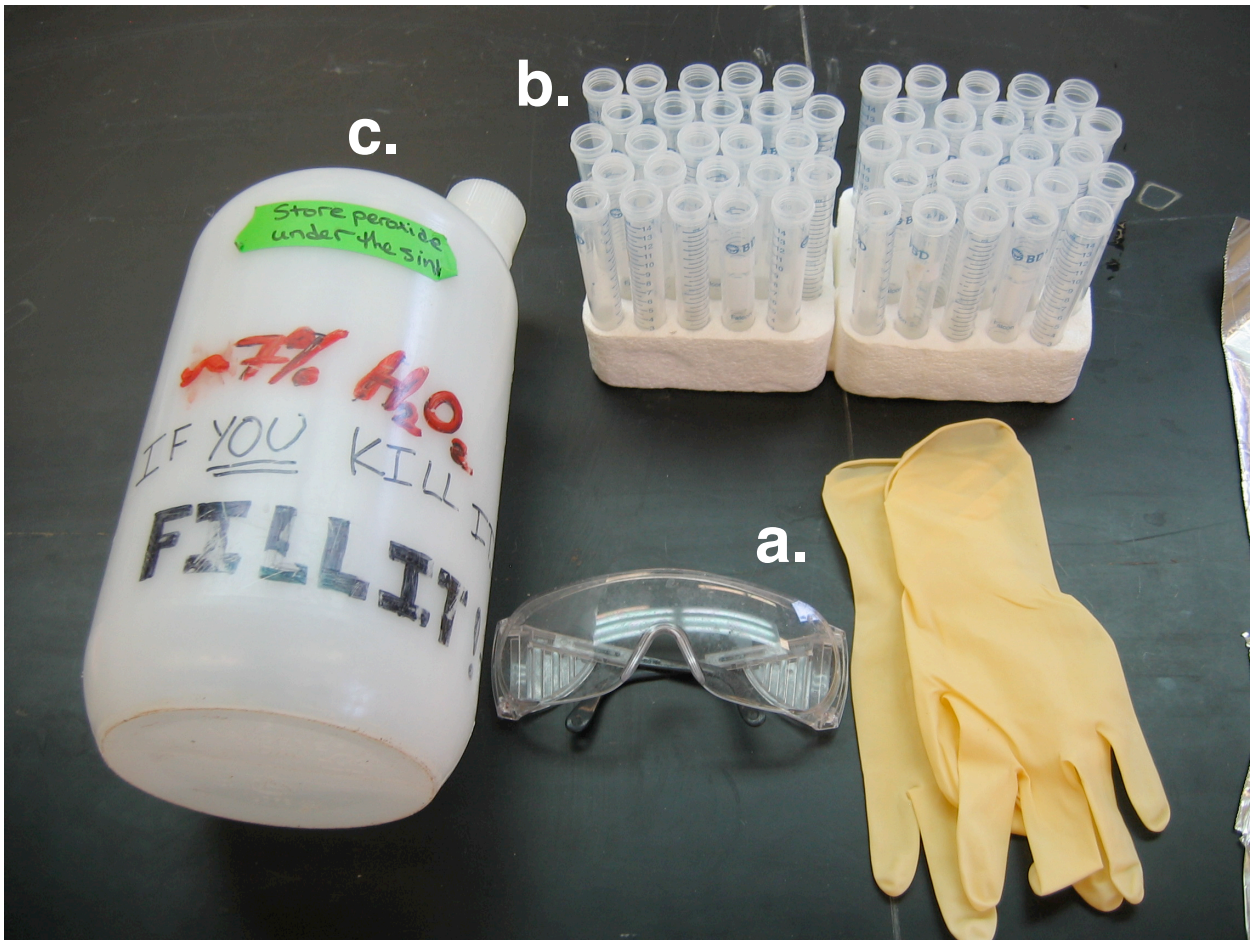
H_2O_2 Treatment

a. Necessary Items

a. Plastic gloves and eye protections

b. Samples in falcon tubes or other container larger than 15ml

c. 7% H_2O_2 63 μm sieve with non-porous bottom to catch sediments



a. Cover sample with 12 mL of ~7% peroxide solution (H_2O_2) (Note that this peroxide solution is a corrosive agent and it is highly advisable to wear gloves and eye protection while using it. Even the weak 7% solution will react unfavorably with your skin).

b. Screw the blue cap on tightly and shake vigorously to help break up the sediment. After shaking, loosen the cap 1 turn to allow the CO_2 to escape, but keep the sampled covered to protect it from material that could fall in.

c. Let the sample soak for a maximum of three days, but long enough to disaggregate the sample material. It is advisable to treat all samples in the same manner. Shake the samples twice a day while they are soaking in peroxide to stir up the solution, which help the reaction break up the sediment. Also check on them to make sure they haven't exploded or bubbled over.

d. When the samples are done soaking, poor off pour off whatever excess peroxide you can **without** losing any sample.

e. If you have to make new H_2O_2 solution, remember that $M_1V_1=M_2V_2$.

a. 35% H_2O_2 * X ml = 7% H_2O_2 * (amount of desired 7% H_2O_2 when you are finished)

b. Solve for X and subtract this from the final volume that you desire.

c. For example: 35% * X = 7% * 3000 ml

d. X = 600 ml so add 600 ml of 35% H_2O_2 to 2400 ml of DI H_2O to get a 7% solution of H_2O_2.

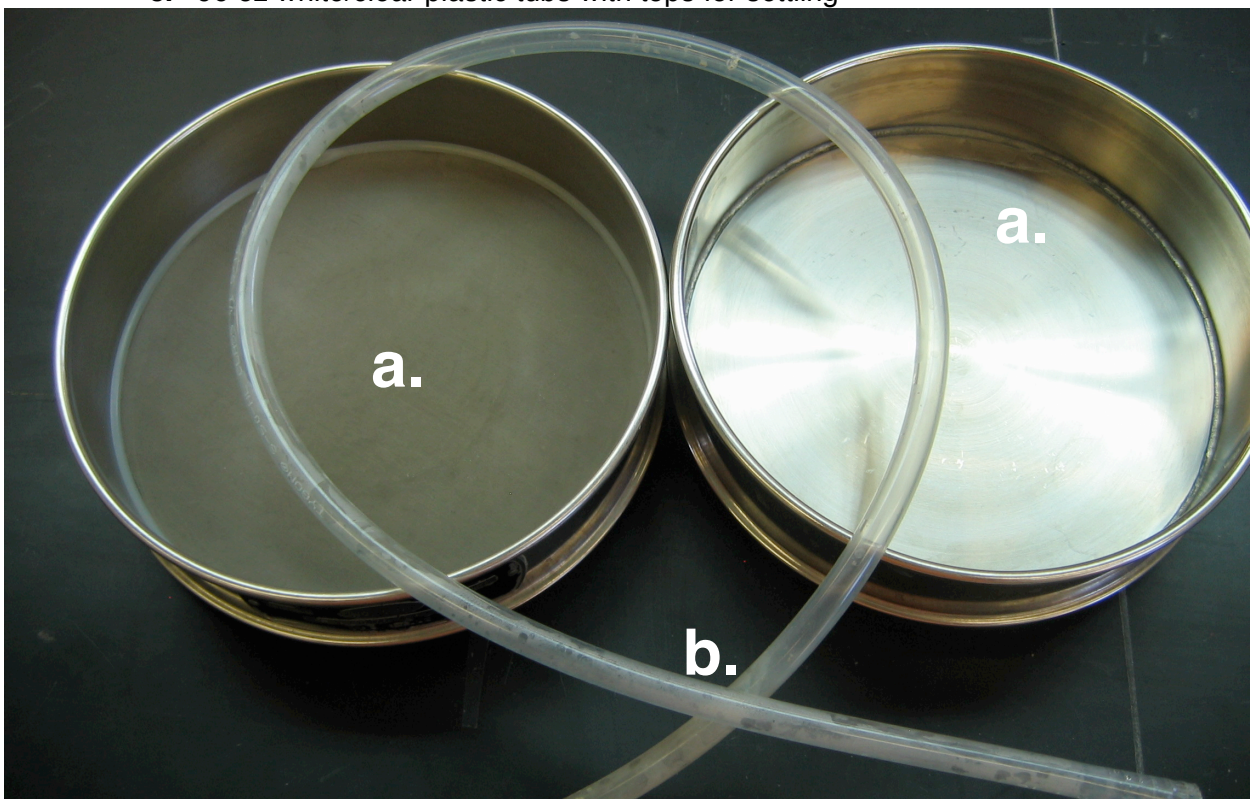
4) Sieving

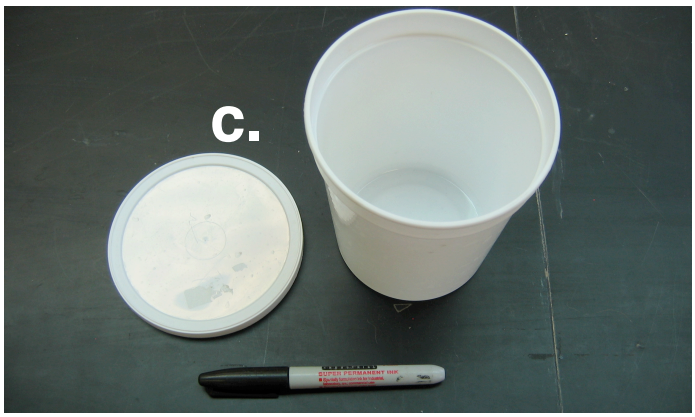
a. Necessary Items

a. 63 μm sieve with solid bottom to catch sediments

b. Rubber tubing attached to faucet for spraying sediments through sieve

c. 96 oz white/clear plastic tubs with tops for settling





b. Turn on the faucet so that a slow, but steady stream of water is flowing out of the surgical tube attached to the nozzle. Do this away from the sieve so that you do not add unnecessary water to the sample that you will have to decant later.

c. Pour the sample and whatever excess H_2O_2 remains out onto a large diameter 63μm sieve that is nested into a solid sieve bottom (**you must catch all of the water that passes through and save it**).

d. Use the water to rinse out the falcon tube that the sediment was disaggregated in onto the sieve **(NO SEDIMENT SHOULD EVER GO DOWN THE DRAIN)**.

e. Place your finger over the end of the surgical tube that the water is flowing from to create a semi-high pressure spray and use it to wash the sediments through the sieve into the sieve bottom. If you are going to be doing this all day, it is advisable to wear rubber gloves so the constant water jet doesn't ruin your hands. Do not spray the sediment so hard that you cause any to splash out of the sieve. Also be careful not to spray so hard that you break apart shell material. **PAY ATTENTION TO THE VOLUME OF WATER IN THE SIEVE BOTTOM SO THAT YOU DO NOT OVERFLOW THE CONTAINER**.

f. Once the sieve bottom is filled with water, or the fine portion of the sediment has all passed through the sieve, remove the sieve from the sieve bottom and set it aside. Pour

the contents of the sieve bottom into a ~96 oz. plastic container. If this represents all of the fine-grained sediment, rinse out the sieve bottom and pour the rinse into the bucket, place the lid on the bucket, put a pre-made label on the bucket, and set it aside on a rollaway cart to sit overnight so that the sediment can settle out of suspension to the bottom. If there is still fine-grain sediment in the sieve, continue sieving until it has all passed through. If necessary, expand into another bucket (this shouldn't usually happen) so that all of the fine-portion maybe preserved.

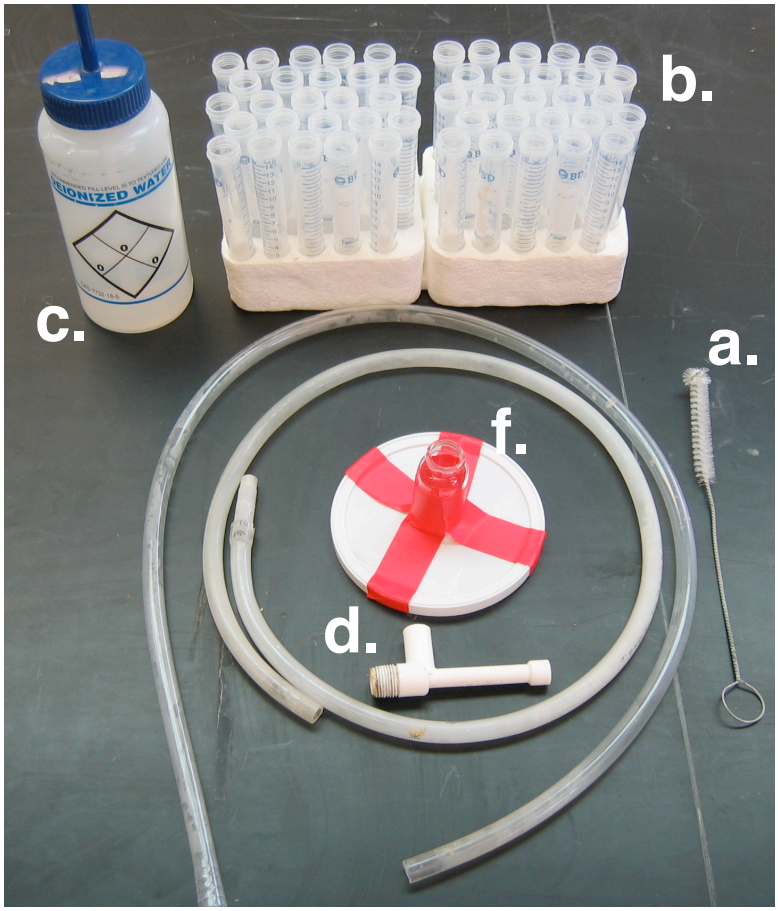
g. Transfer the label from the Falcon tube to the top of the settling bucket. Make a new lable if more than one bucket is used. To make the label transfer easy, **leave a tab on the label when you initially tape it to the Falcon tube**

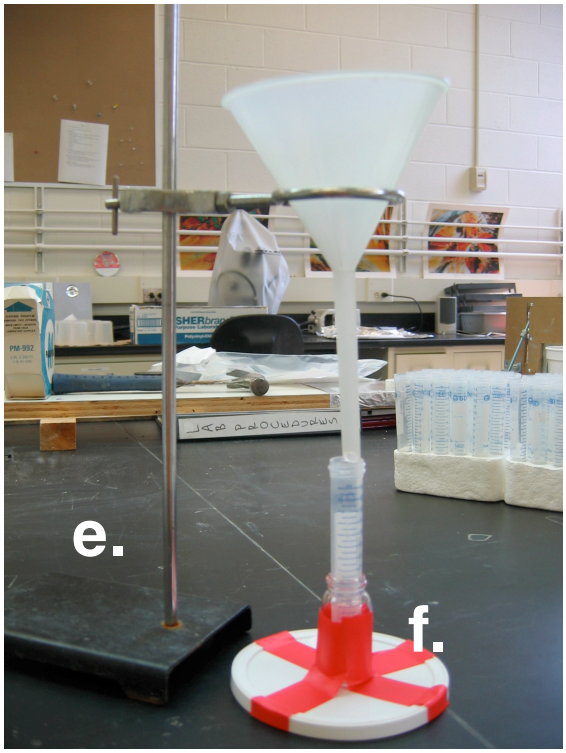
h. Allow the fine-grained portion to settle in the buckets for at least **24 hours** to ensure that all possible material has settled out.

5) Decanting

a. Necessary Items

- **a.** Bottlebrush
- **b.** Cleaned Falcon tubes
- **c.** DI H_2O
- **d.** Hydro vacuum with associated tubing
- **e.** Funnel and stand
- **f.** 15 ml Falcon tube support stand





b. Before you decant the settled sample and pour it back into the Falcon tube from which it came, you must wash the Falcon tubes so that there isn't any unsieved material in them. Use a bottlebrush and DI water to clean the Falcon tubes.

c. After the sample has settled, you will need to collect the carbonate from the bottom of the bucket. To do this, use the hydro vacuum attached to the faucet to suck off the supernatant. Be careful to suck up as little of the carbonate as possible during this step! However, **be aware that you will lose some sediment, just try to minimize the loss**. Remove the supernatant to the lowest level possible without loosing a significant amount of sediment. The point here is that you want to remove enough water so that all the carbonate slurry can fit in the 15 mL Falcon tube when you finish. Some material will be lost, but you must try to minimize this. If you accidentally stir up material during this step, set the bucket aside and allow it to settle for an additional 24 hours.

d. Before pouring the carbonate slurry from the settling bucket back into the Falcon tube, remove the label from the top of the settling buck and tape it back on to the Falcon tube.

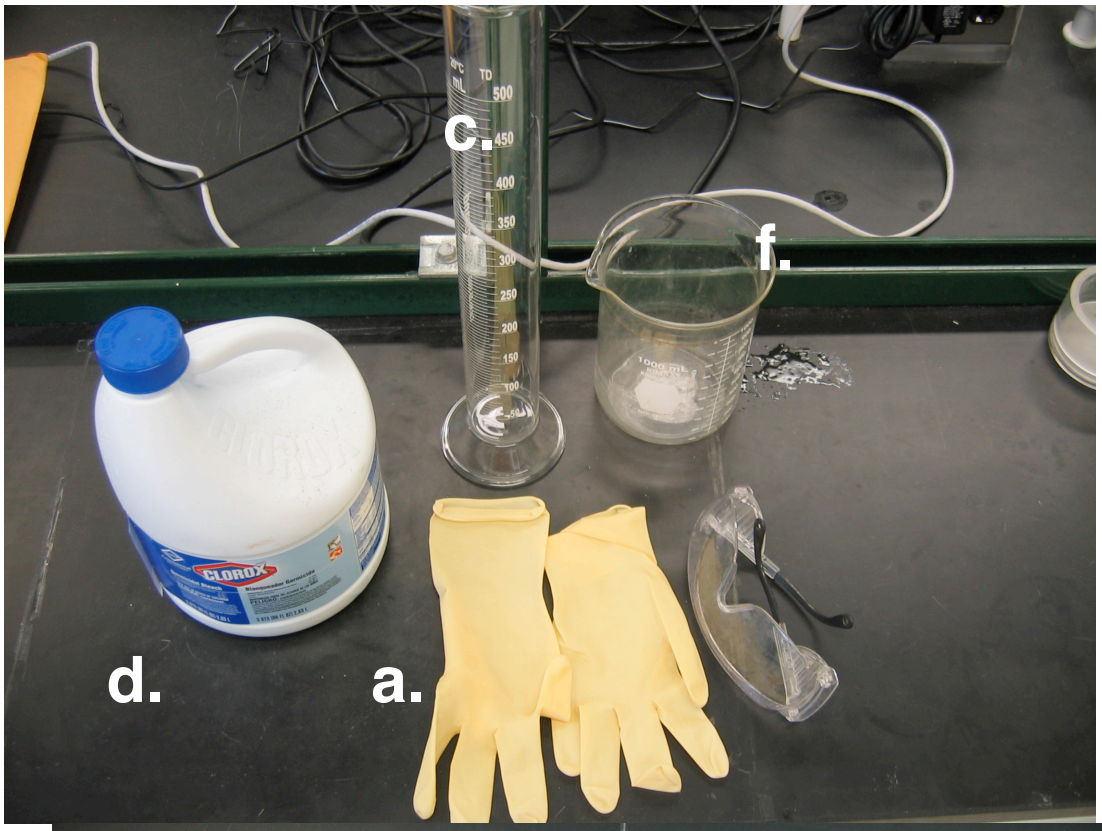
e. Once you have removed all the water that you can safely, you will transfer the material via a funnel back into the Falcon tube. Use the funnel stand and the Falcon tube support for this step. Put the Falcon tube in the support (the makeshift stand is a bucket top with a glass vial taped to it) and position it under the funnel. Swirl the water in the bucket to mix up the remaining sediment. Carefully pour the slurry into the funnel and into the Falcon tube. **BE SURE THAT THE FUNNEL IS CORRECTLY POSITIONED OVER THE FALCON TUBE.** Use a squirt bottle with DI H_2O to rinse any remaining sediment out of the bucket. Also rinse the funnel into the Falcon tube to get all material. If you mess up and have too much water in the bucket, you can centrifuge the Falcon tube for 60 seconds to settle the material to the bottom of the Falcon tube, then poor off the water without loosing any sediment and then repeat the transfer process above.

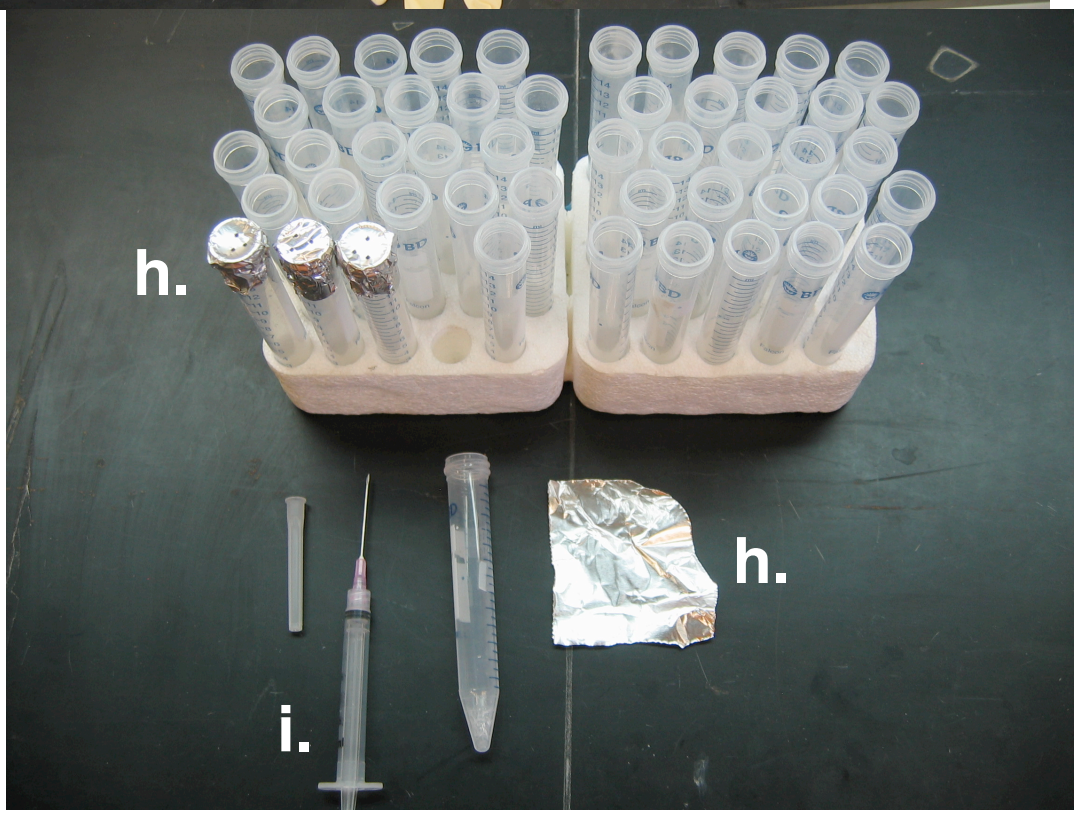
f. After you have transferred all of the samples back to the Falcon tubes, bring them upstairs to the centrifuge in SRCC 416. Centrifuge the samples at 4000 rpms for ~45 seconds. If you do not know how to use the centrifuge, **STOP** find someone to show you! After this, all the material should be at the bottom of the Falcon tube. Poor off the water.

7) Bleaching

a. Necessary Items

a. Gloves and eye protection
b. Lab coat
c. 500 or 1000 ml beaker
d. Regular strength household bleach
e. DI H_2O
f. Container for 50% bleach solution
g. Metal spatula
h. Aluminum foil
i. Needle





b. Bleach is nasty stuff and it WILL ruin your clothing and skin. Make sure to wear protective clothing (eye glasses, gloves, and a lab coat).

c. Mix a solution of 50% bleach %50 DI H_2O. Poor 10-12 mL of the bleach solution into each of the Falcon tubes and let them soak for 6 to 8 hours, no more, no less.

d. Make sure that the sample is totally disaggregated in the bleach solution. This may require you stirring the sample with a metal spatula. **CLEAN THE SPATULA BETWEEN SAMPLES TO AVOID CONTAMINATION**.

e. Decant the bleach solution off after centrifuging in the same manner as described above.

f. After decanting the bleach off the samples, fill each Falcon tube with 10-12 mL of DI H_2O and shake the samples to mix the sediment up. Centrifuge and decant the samples to remove the DI H_2O as described above. Repeat this rinsing process three times. After the third rinse and decanting, leave the Falcon tube empty with only the sediment remaining (there will be a tiny bit of H_2O left, this is ok).

g. Leave the blue Falcon tube cap off each sample after the final rinse. Cover the Falcon tube with a piece of aluminum foil. Poke three (3) holes in the aluminum foil covering each Falcon tube

h. Place the aluminum foil covered falcon tubes in a freezer for 8-10 hours (overnight).

8) Freeze Drying

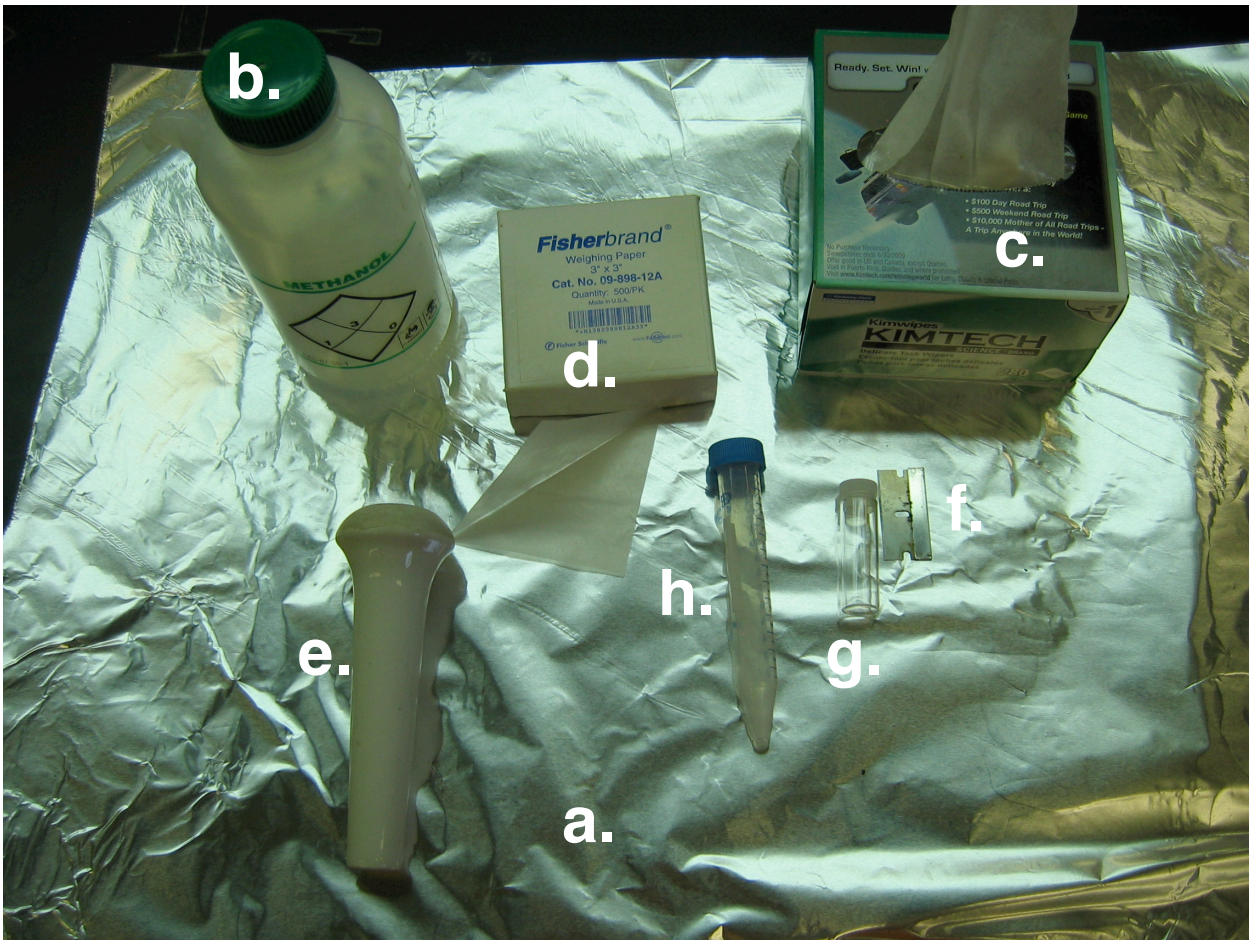
A. YOU MUST BE TRAINED ON THE FREEZE DRIER BEFORE YOU CAN USE IT.

b. Freeze dry the sample for 36 hours.

8) Chopping (Homogenizing)

a. Necessary Items

a. An aluminum foil working surface
b. Methanol in a squirt bottle
c. Kimwipes
d. Weighing papers
e. Pestle
f. A rectangular razor blade
g. Glass shell vials
h. Sample



b. First prepare the working surface by spraying it with methanol and wiping it down with a Kimwipe.

c. Select the sample you are going to homogenize

d. Remove the label from the Falcon tube and place it on the glass shell vial that you are going to which you are going to transfer your sample. If necessary, place additional tape on the shell vial to secure the label.

e. Take a piece of weighing paper and fold it in half diagonally (this will be your funnel to transfer the sample into the glass shell vial.

f. Take the Falcon tube with the sample and breath on it near the bottom to reduce static electricity. Take the tube and turn it upside down in one swift motion and place it on the weighing paper quickly and carefully, not spilling any material. Take the pestle and tap the Falcon tube several times to jar loose any material that is stuck in the tube. If necessary, use a metal spatula to dislodge additional sample. **CLEAN THE SPATULA BETWEEN SAMPLES**.

g. Lift up the Falcon tube and you should have a small pile of carbonate. In some cases you may also have some fibrous material left over from the organic fraction, this ok and it should be incorporated into the final homogenized sample. Take the razor blade and carefully chop up the carbonate and organic material. You may choose to crush the carbonate material first with the pestle to break it up. Once the material is a fine powder with no major chunks, you are ready to transfer it into the glass shell vial. Pick up the corners of the weighing paper so that the carbonate accumulates along the crease in the

middle of the paper. Position the paper over the glass shell vial and gently tap the paper so the carbonate empties into the vial. Cap the vial tightly. Throw away the weighing paper once the sample has been transferred. **NEVER USE THE SAME WEIGHING PAPER TWICE.**

h. Finally, between each sample you must clean the workspace. If you accidentally spill some sample, a clean workspace will allow you to pick it up and continue working with the sample because you know it wasn't contaminated.

i. Label the box containing the shell vials with the samples. Label the top, front, and side of the box.